Bioindication of Ozone Using Milkweed Plants in Southern Ontario

By

Vanessa Redwing-Shaw, B.Sc.

A Thesis
submitted to the Department of Earth Sciences
in partial fulfillment of the requirements
for the degree of
Master of Science

Brock University
St. Catharines, Ontario

Vanessa Redwing-Shaw © 2009

Abstract:

Ambient (O_3) ozone concentrations were compared to ozone damage on milkweed plants to determine if there was a correlation. Eight survey sites of at least 100 plants each were located within 5 kilometers of Air Quality Index (AQI) stations in southern Ontario. Sites were visited nine times from June-September (2007) and milkweed leaves from 75 plants were assessed using methods pioneered in the United States. Ambient O_3 results were calculated into SUM65, seasonal cumulative O_3, and total O_3. The O_3 exposure indices SUM65 and cumulative O_3 were tested statistically to determine which index is biologically relevant to milkweed as an O_3 damage indicator species. The milkweed damage indices were incidence of leaves damaged per plant, incidence of plants damaged per site, and total O_3. The incidence of plants injured per site was the best damage parameter with an $F(1,28)=17.37$, $p=0.0003$ for SUM65 and $F(1,28)=7.5$, $p=0.0106$ for cumulative O_3.. Milkweed plants showed quantifiable ozone damage with minimal spatial differences in damage and thus have potential use as a biomonitor species in southern Ontario.

Acknowledgments:

The completion of this master's thesis would not have been possible without the guidance of many people. First I would like to thank my supervisor and mentor Dr. Daniel McCarthy, without his leadership this work would not be possible. Dr. McCarthy and I have worked together since 2003 and in that time I have been gently guided through two degrees in the Earth Sciences Department. His knowledge, experience and unique perspective have provided me with valuable insight over the years and under his direction I have developed into an independent, critical thinker. Secondly, I would like to thank Dr. Uwe Brand who brought about a wonderful balance to my supervisory committee. Dr. Brand not only supported all of my endeavors over the years, but was always available for discussions on career choices and life experiences. I have been lucky to work with Dr. Brand and am thankful for his advice both technical and personal.

I would also like to thank the Earth Science Department, especially Diane Gadoury, Frank Fueten, Rick Cheel, John Menzies, Astride Silis, Howard Melville, and Holly Arnold for their support and patience over the years.

I must thank my family for bringing me to Brock University, for being so wonderful and for providing constant encouragement without which the challenge of successfully starting school again may not have been met.

Finally, I would like to thank my husband, Josh Shaw, who has nearly as much as I do invested in this thesis. I was lucky to have him as my official research assistant during the field season, editor during the writing season and general advisor throughout the whole process. Josh provided this help in addition to giving me support when I needed it and constant encouragement to get the job done.

In addition, this research would not have been possible without the assistance of the Ministry of the Environment especially Mrs. Laura Fiore and Bob Emerson, the financial support of Grand River Post Secondary Educational Office, Brock University Graduate Studies, Harrison-Thompson Bursary Trust, National Aboriginal Achievement Foundation, Petro-Canada, Indigenous Health Research and Development Program, Casino Rama, Geological Association of Canada.

Table of Contents

List of Figures:

List of Tables:

Chapter 1: Bioindication of Ozone using Milkweed Plants

1.1 Introduction

Ozone is a gas that is ubiquitous globally and can be useful or harmful depending on where in the atmosphere it is found (US EPA, 1996). When O_3 is formed in the stratosphere it forms the protective O_3 layer, which makes Earth a habitable planet by blocking harmful ultra-violet rays. However, when O_3 is formed at ground-level its production is fueled by anthropogenic emissions. Ozone is of interest because it is known to have adverse effects on the environment and the health of its inhabitants including certain sensitive plant species. In laboratory animals it has been found that long term exposure to O_3 can result in damage to the lung, heart, liver, immune system and brain, increasing occurrence of tumor formation in these areas (Mustafa, 1990; Bell, McDermott, Zeger, Samet, Dominici, 2004). In response to O_3 exposure, some plants have been found to exhibit tissue and cellular damage, which can lead to premature cell death (Iqbal, Abdin, Mahmooduzzafar, Yunus & Agrawal, 1996; Pell, Schlagnhaufer & Arteca, 1997; Schraudner, Langebartels & Sanderman, 1997).

In Canada O_3 is legally classed as a toxic chemical that poses a known danger to human health as defined by the Canadian Environmental Protection Act (CEPA, 1999), the environmental legislation established in 1999. The Canadian government through CEPA is charged with monitoring atmospheric O_3 using air quality stations. At the federal level the National Air Pollution Surveillance (NAPS) Network operates stations nationwide and provincially there is the Air Quality Index (AQI) network of stations. In Ontario, the Ministry of the Environment Environmental Monitoring and Reporting Branch has a network of 40 fixed sensors that are used to monitor ambient air quality (MOE). The determination of air standards, monitoring and regulation of Ontario's air quality by the MOE is based on the current body of scientific knowledge and MOE research (MOE). The monitoring network and site specific studies involve the use of mobile and permanent sensors, bioindication studies and partnerships with various universities and industries, which provide data that can be used to track spatial and temporal changes in ground-level O_3 across the province. Once the data have undergone quality control analysis the information is compiled into a report that is released to the public on a yearly basis (e.g., Ministry of the Environment Air Quality in Ontario 2005) (MOE, 2006).

Air Quality Index (AQI) stations record levels of six atmospheric pollutants, which are used to calculate an Air Quality Index number attributing a value for each hour of each day that indicates a measure of air quality (MOE). The AQI is calculated hourly based on these data, a high value (51-100) indicates poor air quality and a low number (<25) indicates good air quality (MOE). From the data collected an Air Quality Health Index (AQHI) is also calculated indicating the level of heath risk due to air quality each day (MOE). This index is currently being tested in a pilot program in Ontario. The 40 stations comprise an efficient network for pollutant monitoring and have accurately characterized baseline O_3 levels in the past. In general, fixed monitoring stations provide a precise time-sensitive overview of geographical variation in O_3 levels. However, unlike point source pollutants O_3 tends to cover large areas with varying levels of uniformity and the resulting data are often extrapolated or interpolated regionally (US EPA, 2006).

Ozone monitoring stations are expensive to build and require maintenance making widespread local monitoring difficult (Manning, Krupa, Bergweiler & Nelson, 1996; Chappelka *et al.*, 1997; Manning, 2003; US EPA, 2006). The conditions for O_3 development vary seasonally, geographically, and are highly sensitive to regional climate conditions, which affect overall O_3 production. Consequently fine-scale differences in O_3 levels may go undetected in areas where microclimatic conditions vary. It is in such areas that biomonitoring has considerable potential as a way to quantify O_3 exposure and add to our knowledge of regional variability (Manning, 2003; US EPA, 2006). It is for this reason that use of cost-effective, widespread indicator plants as a means of characterizing O_3 pollution should be examined.

Elevated O_3 levels have historically been documented using the network of AQI sensors, and bio-indication using higher plants has been explored as a means of expanding the amount of information gathered (MOE). Plants such as milkweed (*Asclepias*) are sensitive to O_3 and have been used as bio-indicators due to their distinctive and readily identifiable injury from O_3 exposure, as demonstrated by studies in the United States (Chappelka, Renfro, Somers & Nash, 1997; Skelly, Ferdinand, Savage, Jagodzinski & Mulik, 2000; Yuska *et al.*, 2003; Bennett, Jepsen & Roth, 2006; Davis & Orendovici, 2006; Souza, Neufeld, Chappelka, Burkey & Davison, 2006). That body of work has used controlled studies to establish O_3 damage thresholds for milkweed plants, which are known to occur above approximately 60 ppb and has established that milkweed has considerable utility as a bio-indicator of ground-level O_3 (Neufeld, Renfro, Hacker & Silsbee, 1992; Chappelka *et a*l., 1997; Pell *et al.*, 1999; Gunthardt-Goerg, McQuattie, Maurer & Frey, 2000; Kouterick *et al.*, 2000). Data from controlled studies are useful, however, it is still imperative to examine O_3 injury in the natural growing habitat, and there are few studies recording milkweed injury, O_3 levels and environmental variables *in situ* (Chappelka *et al.*, 1997; Somers, Chappelka, Rosseau & Renfro, 1998; Vollenweider, Ottiger & Gunthardt-Goerg, 2003; Yuska *et al.*, 2003; Schaub *et al.*, 2005; Bennett *et al.*, 2006). Unfortunately milkweed has seen little use as a biomonitor in Canada. However, using milkweed plants as indicators of O_3 exposure could improve our ability to track spatial and temporal changes in ground-level O_3 in Ontario.

Ontario often has Canada's highest O_3 levels, (Figure 1.1) (Toronto Public Health, 2005; Yap, Reid, De Brou & Bloxam, 2005; MOE, 2007). Southern Ontario is particularly prone to high O_3 levels because it not only has its own primary pollutants, but it receives pollutants from the United States Midwest and the Ohio Valley. The financial and biological significance of this pollution regime is hard to quantify, various estimates suggest that direct and indirect impacts of ground-level ozone and fine particulate matter in Canada could approach \$9.6 billion per year (Yap *et al.*, 2005). Annually, \$5.2 billion in damages is attributed to transboundary air pollution with most of this impact felt in south central and south western Ontario (US EPA, 2004; Yap *et al.*, 2005; MOE). Clearly, it is imperative that ground-level O_3 be monitored in the most effective ways possible so as to develop a clear understanding of the spatial and temporal differences in ground-level O_3 in Ontario. Accordingly, this thesis will attempt to explore how best to use milkweed as a bio-indicator of ground-level O_3, testing the utility of various indices, and documenting the response across an ozone-rich area in Ontario. Using milkweed will provide one of the first known systematic surveys of O_3 impact on milkweed plants with geographic variability in southern Ontario.

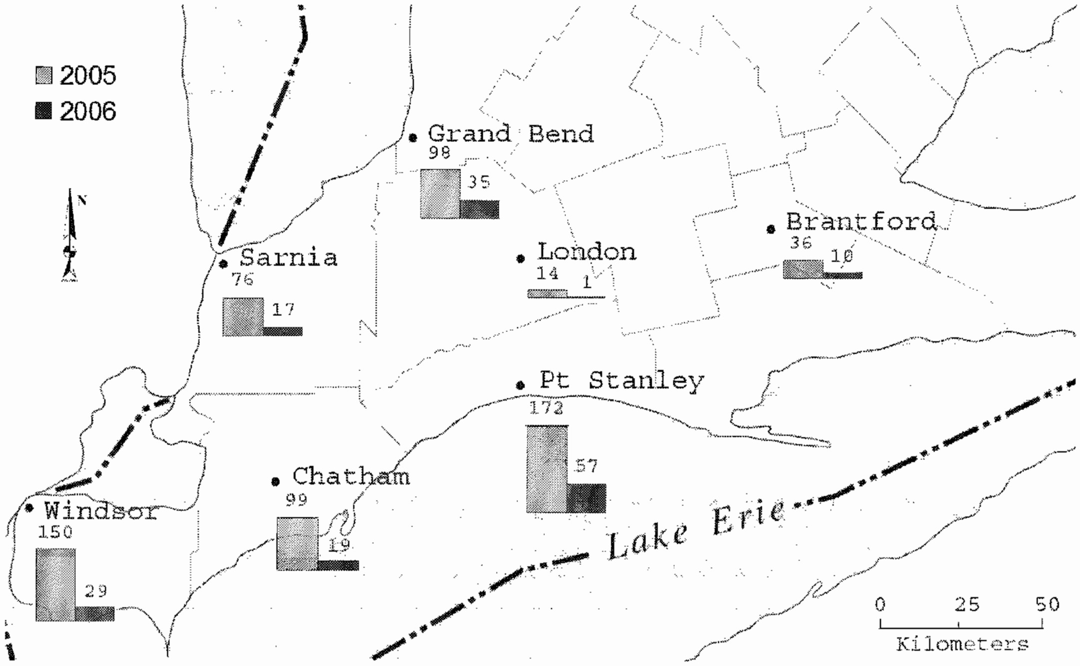


Figure 1.1: Locations of 7 Air Quality Index stations in southern Ontario, bars indicate the 1-hour exceedances over 80 ppb which is the current (2008) air quality standard. The 1-hour exceedances for Simcoe are not available (MOE, 2007; Map base supplied Brock University Map Library).

1.2 Approach and Research Objectives

The primary aim of this thesis is to provide a detailed demonstration and testing of the use of milkweed plants as bio-indicators of ground-level O_3 in southern Ontario. It will review previous O_3 monitoring studies using milkweed plants, meteorological variables affecting O_3 production and their effect on plant uptake, and will review how foliar O_3 damage occurs. By correlating known O_3 levels with plant damage the accuracy, consistency and variability of milkweed as an indicator of O_3 pollution will be tested. The field component will attempt to address the following three objectives through assessing correlation and not causality:

- Systematically document milkweed foliar damage in O_3 rich areas of southern Ontario

- Statistically explore the link between foliar damage and O_3 exposure in southern Ontario, while identifying which damage and exposure indices are most appropriate for milkweed studies
- Characterize the relationship that exists between meteorological variables and ozone-induced foliar damage in southern Ontario.

The hypotheses being tested are that *there is an identifiable relationship between O_3 exposure and milkweed O_3 damage*; *there is an effect of total daily precipitation on milkweed O_3 damage; that there is an effect of mean daily air temperature on milkweed O_3 damage* and *that there is measurable, geographic (site) differences in O_3 related damage to milkweed.*

Chapter 2: Ozone and Ozone Monitoring

2.1 Tropospheric and Stratospheric Ozone Formation

Ozone is a secondary product of photochemical, or sunlight-induced, reactions and occurs in both the troposphere and the stratosphere (McKee, 1994; US EPA, 2006). The photochemical reactions that create and also destroy O_3 will be different depending on whether they are occurring in the troposphere or stratosphere (McKee, 1994). In addition the function of the O_3 is different depending on its location (McKee, 1994; US EPA, 1996). In the stratosphere O_3 blocks the passage of ultraviolet rays allowing for life on Earth to exist (McKee, 1994). When O_3 is at ground level there is a potentially negative impact on the inhabitants of Earth, possibly causing injury to plants, trees, animals, humans and property, otherwise in the stratosphere it functions as a UV-blocker (McKee, 1994; US EPA, 2006).

Stratospheric O_3 formation and destruction is relatively straightforward: molecular oxygen (O_2) is broken down in the presence of sunlight producing two oxygen (O) atoms and during this process ultraviolet rays are absorbed (US EPA, 2006). The separate oxygen atoms may either rejoin with a single oxygen to reform O_2 or it may go with a molecular oxygen (O_2) to form ozone (O_3) (McKee, 1994; US EPA, 2006), (Table 2.1). The O_3 layer is preserved because the process of formation and destruction is continually taking place. Ozone produced in the stratosphere absorbs much of the harmful ultraviolet radiation produced from the sun and is essential to life on Earth (McKee, 1994).

Tropospheric O_3 formation follows a complex set of reactions when combined with the pollutants and emissions found at ground-level (McKee, 1994; US EPA, 2006). In ground-level O_3 production a reaction between nitrogen oxides (NO_x), volatile organic compounds (V.O.C.) and oxygen (O_2) occurs with sunlight acting as the catalyst (McKee, 1994; US EPA, 2006). The amount of O_3 formed naturally at ground level is minimal (McKee, 1994). The majority of toxic ground-level O_3 produced is due to elevated levels of anthropogenic and automotive emissions with coal-fired power plants contributing a major supply of nitrogen oxides and volatile organic compounds in the lower atmosphere (McKee, 1994; US EPA, 2006).

Table 2.1: The photochemical reactions that produce and destroy ozone in the stratosphere. Source: Atmospheric Ozone and Data Resources (NASA).

- Stratospheric Ozone Production:

$$O_2 + \text{UV light} \rightarrow 2\,O$$

$$O + O_2 \rightarrow O_3$$

- Stratospheric Ozone Destruction:

$$O_3 + \text{UV, visible light} \rightarrow O + O_2$$

Nitrogen oxides (NO_x) are a principle component in O_3 formation. This term is used to indicate when both nitric oxide (NO) and nitrogen dioxide (NO_2) are present (US EPA, 2006). NO_x are found naturally in the atmosphere from sources such as forest fires, soil processes and lightning (US EPA, 2006). However, most of the NO_x in the atmosphere is the product of combustion reactions from motor vehicles, power plants and industrial processes (US EPA, 2006).

Volatile organic compounds (VOC) are readily vaporized, carbon-based compounds that occur in the atmosphere from natural and anthropogenic sources (McKee, 1994). In their natural form VOC are emitted by plants, cattle, wetlands and farming activity (Climate Change, 2001). In their anthropogenic form VOC are vaporized from a great many products some of which include solvents, disinfectants, formaldehyde, cigarette smoke, fuel, paint thinner, pesticides, adhesives. (US EPA, 2006). Volatile organic compounds are important in O_3 production because V.O.C. can produce nitrogen dioxide without using the O_3 molecule as a reactant resulting in more O_3 production and less O_3 destruction (McKee, 1994).

In tropospheric O_3 production there are dozens of complex reactions that might occur. The general explanation for this process is as follows: ultraviolet rays split nitrogen dioxide (NO_2) resulting in nitric oxide (NO) and atomic oxygen (O). The oxygen atom can then go with molecular oxygen (O_2) to produce O_3,which can react with nitric oxide (NO) producing nitrogen dioxide (NO_2) and molecular oxygen (O_2), (Table 2.2) (NASA: Earth Observatory). These reactions are photochemical in nature the local meteorological conditions will in part determine how much O_3 is produced.

Table 2.2: A simplified form of the photochemical reactions of tropospheric ozone formation and destruction. Source: (NASA, Earth Observatory).

- Tropospheric Ozone Production:

$$NO_2 + \text{UV energy} \rightarrow NO + O$$

$$O + O_2 \rightarrow O_3$$

$$NO + O_3 \rightarrow NO_2 + O_2$$

$$NO_x + VOC + \text{UV energy} \rightarrow O_3 + NO_2 + \text{other chemical species}$$

2.2 Meteorological Conditions that Affect Ozone Formation

The amount of O_3 produced in the troposphere is not only influenced by emissions, but also by the meteorological conditions that occur at ground level (McKee, 1994; US EPA, 2006). The most important factors in O_3 production and subsequent uptake by plants are sun availability, mean daily air temperature, wind speed and total daily precipitation (US EPA, 2006).

It is known that air temperature, O_3 production and volatilization of organic compounds are positively correlated (Kelly, Ferman & Wolff 1986; Kelly and Gunst, 1990; US EPA, 1996). Consequently, in eastern North America O_3 production is more prevalent in the summer months when solar radiation and air temperature are high (McKee, 1994; US EPA, 1996). In Ontario the summer high temperature and high pressure are associated with the south-west winds that bring NO_x-rich air from the central US. The increased air temperatures of the summer season that are associated with high pressure meteorological systems, which will act to reduce wind speed allowing for O_3 buildup (McCurdy, 1994; US EPA, 1996). Sinking of air is associated with a high pressure system and produces little cloud cover and low wind speed, resulting in a stagnant O_3 blanket, which becomes concentrated, and can affect an area of hundreds of square kilometers (McCurdy, 1994; McKee, 1994; US EPA, 1996). Ozone can accumulate in urban centers, if the wind speed is increased the O_3 will be transported and an extension of the O_3 plume can be found downwind resulting in elevated O_3 levels and widespread exposure (McCurdy, 1994; US EPA, 2006). Stagnant

O_3 plumes can be produced in the summer months also due to an increased demand for electricity during heat waves resulting in the burning of more fossil fuels, which will release primary pollutants into the air facilitating more O_3 production (Environment Canada, 2007).

Ground-level O_3 is elevated due to anthropogenic emissions and there are few places on Earth that humans have not affected, it is difficult to determine background, or naturally occurring concentrations of O_3. Health Canada in 1999 measured background values at fifteen remote sites within Canada and determined the range to be 25-40 ppb in the summer months (McKendry, 2004). A more recent study of North America found background O_3 levels to be between 24-45 ppb (Fiore, Jacob, Liu, Yantosca, Fairlie & Li, 2003). It should be understood that background levels of O_3 in other countries have been found to be on the rise (Nolle, Ellul, Heinrich & Gusten, 2002).

2.3 Monitoring Using Mechanical Sensors

In North America, O_3 and related atmospheric pollutants can be monitored by a combination of permanent, mechanical sensors and mobile, parameter-specific units. These are maintained by the governments of Canada and the U.S., universities and private industry. Typically, in Ontario sites closest to the international border receive transboundary pollution and for this reason international O_3 monitoring takes place (MOE, 2006).

Air pollutants in Canada are monitored by two networks established in the 1970s with the advent of the first chemiluminescent machines (continuous air samplers) (Pearson, 1992; MOE, 2007). Nationally, Canada maintains the NAPS and the CAPMoN network, which is comprised of 18 O_3 monitors. Provincially there is the Air Quality Index (AQI) system, which includes 40 sensors. The AQI stations take in continuous air resulting in air quality raw data on ambient levels of O_3, nitrogen oxides, carbon monoxide, volatile organic compounds, sulfur dioxide, total reduced sulfur compounds and fine particulate matter. The O_3 data for this study were retrieved from the AQI network. Additional information on the CAPMoN network can be found at: http://www.msc-smc.ec.gc.ca/capmon/index_e.cfm. In 2006 the 40 fixed sensors used to monitor O_3 at the AQI sites. These are distributed across Ontario with 12 near the Canada-U.S. border and in urban areas. An unknown number of mobile and study-specific sensors were used by university and industry-based researchers in Ontario (MOE, 2007). From the hourly data generated by the AQI network two relative air quality index values are calculated based on the highest reading of these pollutants each hour. This includes the Air Quality Index, which indicates the relative quality of air reported in five categories, and the Air Quality Health Index (AQHI), which indicates the relative level of heath risk due to air quality each day calculated from three air pollutants (MOE). The AQHI is currently being tested as a pilot program in Ontario.

The raw data undergoes a quality-assurance and control (QAC) process and the information is compiled by the MOE into an annual report available to the public. These reports make it easy for policy makers to visualize trends in pollutant concentrations and assess if established limits have been reached or exceeded. (MOE, 2006).

Ozone levels are usually measured in Canada at hourly intervals in parts per billion (ppb). They can also be reported as a daily maximum in ppb or calculated into AQI values: a cumulative concentration in ppb, number of hours or days in exceedance of a particular value, percentage of hours in each AQI category, and as the highest one-hour value (MOE, 2006; US EPA, 2006).

Ozone data can also be calculated into additional indices that are useful for understanding pollutant levels (MOE, 2006; US EPA, 2006). There is a large variation in the O_3 levels that areas of southern Ontario are exposed to each year and there are many indices that can be used to characterize the levels. For this study biologically relevant indices are of most interest because they focus on the range of O_3 exposures that are most likely to effect plants. The reporting formats used by Canada and the U.S. are the 12-h average of O_3 levels from 8:00 a.m. to 8:00 p.m., which averages the hourly O_3 data from 0800-2000 hours each day; and the W126 index that assigns a higher value for exposure values above 40 ppb based on ozone-induced plant damage (US EPA, 2006). The SUM indices reflects O_3 concentrations that exceeded the given value. For example SUM60 will give the cumulative value (ppb) for all O_3 exceedances over 60 ppb (Davis and Orendovici, 2006; US EPA, 2006). The N100 will give the total number of hours the O_3 concentration exceeded 100 ppb. This index is only relevant in regions that experience highly elevated O_3 levels (Davis and Orendovici, 2006; US EPA, 2006).

On a national scale the Canada Wide Standard (CWS) of 65 ppb (8-hr average) for O_3 was established in the year 2000. This level was chosen because O_3 is known to cause damage to plants, animals and humans above 65 ppb (U.S. E.PA., 1996). A timeline has been set by which all locations in Ontario are to adhere to the 65 ppb concentration, the goal is to reduce O_3 levels to meet the CWS by 2010 (MOE, 2006). It should be noted that in 2006 all O_3-monitoring AQI sites in Ontario except Thunder Bay exceeded the 65 ppb CWS set by the Council of Ministers of the Environment (MOE, 2006).

In Ontario the current provincial one-hour Ambient Air Quality Criteria (AAQC) for O_3 measures how many hours a site exceeds 80 ppb. The AAQC will remain the current O_3 standard until the CWS comes into effect in 2010. Data collected from permanent AQI sites from 1980-2006 show there has been a slight decrease in the O_3 one-hour maximum concentrations yet the average is still above the AAQC of 80 ppb (MOE, 2007). The annual mean O_3 exposure has seen an increase over the same period rising 27 and 50% in the summer and winter months, respectively (MOE, 2007). In 2006, 35 of the 38 sites that recorded O_3 data exceeded the AAQC of 80 ppb (MOE, 2007).

2.4 Monitoring Using Biological Organisms

Information about pollutant exposure can be collected from living organisms and used in conjunction with the data collected from mechanical sensors to give a greater understanding of the pollutant impact on biological systems. This is referred to as biomonitoring, which can allow for the assessment of ecological changes and possible habitat and biodiversity losses, while establishing a baseline for future studies (Spellerberg, 2005; MOE, 2006). For example when air quality in a particular area is in question and there is no permanent sensor available, researchers may be able use evidence obtained from plants as a way to estimate spatial differences in air quality.

Over the last sixty years the correlation between bio-indicators and known pollutants has become more accurate and many biomonitoring protocols are readily available from the literature (Spellerberg, 2005). Many plant and animal species have been identified as bio-indicators of specific anthropogenic perturbations (Markert, 2003). A bio-indicator species can express physical changes, damage or reduced abundance in the presence of a pollutant indicating pollutant exposure and in some cases this can be quantified (Manning, 1996; Kohut, 2005).

According to the U.S. Environmental Monitoring and Assessment Program there are several criteria that identify a good bio-indicator species (Hunsaker, Carpenter & Messer, 1990; Spellerberg, 2005). The ideal bio-indicator species would exhibit consistent injuries based on the biotic and abiotic changes due to pollution, be taxonomically identifiable and have readily identifiable injuries (Hunsaker *et al.*, 1990). It is important that the injuries due to pollution exposure are not mistaken for injuries due to other stressors, which could confound the results (Hunsaker *et al.*, 1990). In addition the species are usually chosen based on previous studies and should have the lowest monitoring error and the highest correlation to known pollutant levels (Hunsaker *et al.*, 1990). Finally the species should be cost effective to monitor (Hunsaker *et al.*, 1990).

The organisms used for biomonitoring studies can fall into two broad categories. Accumulator species will uptake pollutants, but suffer minimal effects, while response indicator species are those that are physiologically sensitive and exhibit injuries or limited abundance in polluted areas (Bargagli, 1998). The former necessitates chemical analysis, termed active biomonitoring, whereas the latter can be analyzed by observation alone, defined as passive biomonitoring.

In recent years higher plants have been commonly used by researchers and government officials in Canada and the U.S. for environmental biomonitoring studies (Koutrick *et al.*, 2000; MOE, 1989). Pollution contact occurs on the soil and plant surface and is subsequently taken up by the plant, making many higher plants a useful monitoring tool (Bargagli, 1998). Accumulator plants have developed a tolerance to the particular pollutants found in their environment making these species ideal for active biomonitoring (Bargagli, 1998). Some of these plants can produce structural changes due to an increased level of pollutants in order to survive, for example, programmed cell death due to high level O_3 exposure to compartmentalize injury (Bargagli, 1998). Other species of higher plants upon phytotoxic chemical uptake will not act as an accumulator, but a response indicator instead, showing visible damage; and these species are useful for passive monitoring.

Agricultural crops have also commonly been used as indicators of atmospheric pollutant exposure (Pearson, 1992). One of the original large-scale biomonitoring studies took place in the United States in 1980 when the National Crop Loss Assessment Network (NCLAN) was established to determine crop yield loss due to O_3 exposure. Through a variety of field-based open-top chamber experiments that fumigated crops with O_3, the dose: response relationships of several agricultural species was investigated (Pearson, 1992). This study was significant because it was the first of its kind to assess ozone-induced response in crops outside of a laboratory. The main objective of this study was to test if the AAQC of 80 ppb was an appropriate air quality standard for O_3 exposure based on the sensitivity of valuable crops (Pearson, 1992). An assessment of the O_3 sensitivity of agricultural crops in Ontario was conducted through an extensive literature review and analysis of the NCLAN data. From this work 12 Ontario "Crops At Risk" were determined including: white bean, potato, tobacco, tomato, onion, winter wheat, soybean, sweet corn, green snap bean, spinach, turnip and hay (Pearson, 1992). Agriculture is a significant part of Ontario's revenue, the MOE conducts annual foliar assessments on many of the 12 crops at risk to this day. In this biomonitoring assessment the use of crops was instrumental in determining that the 80 ppb criterion for Ontario was sufficient to protect moderately sensitive crops. If the 80 ppb criteria was adhered to the agricultural community of Ontario stood to increase their profits by an average of $39 million annually (1989 CAD) (Pearson, 1992).

Lichens have also been widely used in both passive and active air quality biomonitoring studies for heavy metals and SO_2, but there remains little evidence that lichens are adversely affected by O_3. As early as 1926 terminology to describe the effects of pollutants on lichen distribution had been introduced (Kricke and Loppi, 2002). "Lichen deserts" and "struggle zones" were related to industrial smoke and sulfur dioxide based on observation alone (Kricke and Loppi, 2002). Eventually a system for quantifying lichen abundance was developed for passive monitoring studies. In areas where heavy metals or SO_2 are of interest, lichen species diversity and percent cover can be quantified along a pollution gradient (Kricke and Loppi, 2002). If pollution levels and one or more measures of lichen abundance can be correlated then maps can be developed to provide a spatial estimation of the various zones of pollution (Kricke and Loppi, 2002). Lichens can also act as accumulator species and are useful for active biomonitoring techniques. In this process lichen samples can be taken from a polluted environment and tested for content. Analysis can include detection of heavy metal or pollutant presence, determination of chlorophyll content and physiological degradation attributable to certain atmospheric pollutants (Nimis *et al.*, 2002).

These are but a few examples of the types of biological monitoring that can be used to indicate air quality. It is important when using bio-indicators to carefully select the species and use a combination of plant part assessments and environmental variables in order to accurately characterize the environment (Bargagli, 1998). The pollutant under study should in part dictate which bio-indicator species is used due to differences in plant uptake, sensitivity and growing conditions (Saarela *et al.*, 2005).

The information collected from the mechanical sensors is clearly imperative to monitoring atmospheric pollutants, however, it holds little informative value in terms of the biological response to O_3 exposure (Pearson, 1992). It is for this reason that data are collected from living organisms that are known to show damage due to atmospheric pollutants. This information is useful because in some situations the sensitive plants can act as early warning for less sensitive plants, and for researchers studying the effects of atmospheric pollutants on natural ecosystems, and agriculture among other things.

Chapter 3: Use of Milkweed to Assess Ground-Level Ozone

3.1 Mechanism of Ozone Uptake and its Effect on Milkweed Plants

Milkweed is a plant that is indigenous to North America. It is known to be sensitive to O_3 exposure and its related injuries have often been used since the 1980s to estimate O_3 levels in ambient air (Duchelle and Skelly, 1981; US EPA, 1996).

The plant damage that results from exposure to O_3 is dependent on the following relationship: O_3 concentration that comes into contact with the plant, O_3 uptake by the plant and the meteorological conditions surrounding the plant (Tingey & Taylor, 1982; Pell and Dann, 1991; Kohut, 2005). The contribution of each of these components is known as the triad of injury; the greatest injury will result when each component is most favorable for uptake (Kohut, 2005).

Upon contact there is little or no O_3 absorption through the plant cuticle itself and it is thought that no damage is incurred from cuticular contact alone (US EPA, 2006). Ozone enters plants through the stomata, the portals for gas exchange. Plants open and close stomata in order to optimize respiration. When stomata are closed they provide some protection from further O_3 damage and moisture loss (Tingey & Taylor, 1982; US EPA, 2006).

Open stomata are maintained by turgor pressure in the guard cells, which is determined by water status of the plant, the availability of sunlight and pollutant presence (Iqbal *et al.*, 1996). Gas exchange is in part regulated by the state of the plant's surroundings. During periods of drought, darkness or pollutant stress, the stomata will lose turgor pressure causing the stomata to close. In the case of pollution, this is an example of an avoidance mechanism, which will limit the amount of pollutant that enters the plant interior (Iqbal *et al.*, 1996). Depending on the pollutant concentration the stomata can either open or close in response to O_3 exposure (Iqbal *et al.*, 1996). For example, it is thought that extracellular exposure to low concentrations of O_3 will increase the permeability of cells that provide water to the guard cells, increasing turgidity and opening the stomata (Iqbal *et al.*, 1996). Conversely, under high exposure (>100 ppb) ozone will directly affect the guard cells causing the stomata to remain closed, which reduces gas exchange (Iqbal *et al.*, 1996; Torsethaugen *et al.*, 1999). Due to the regulating function of the stomata, the amount of O_3 inside and outside of the plant is not necessarily equal (US EPA, 2006). It is important to understand the microenvironmental conditions surrounding the bio-indicator plant such as moisture will play a significant role in O_3 uptake.

Once O_3 has entered the extracellular space there are several changes in metabolism and plant responses that can take place (Tingey & Taylor, 1982; US EPA, 2006). It is within the plant interior in the intracellular spaces that the most relevant O_3 injury occurs and the plant's response to O_3 exposure can be one of avoidance or tolerance. It is this reaction that can incur ozone injury (Tingey & Taylor, 1982; Pell *et al.*, 1997; Schraudner *et al.*, 1997). It is important to note that the complete sequence of response events is not completely understood, and there is extensive research being conducted on the subject (Rao, Koch and Davis, 2000; US EPA, 2006).

3.2 Plant Response to Ozone Exposure

Ozone is an oxidant, which means it can "steal" electrons from neighboring molecules known as free radicals, which are a type of reactive oxygen species (ROS) (Iqbal *et al.*, 1996). When O_3 exposure produces a free radical by stealing electrons, that molecule will become highly unstable and may take electrons from its neighboring cells, which can lead to cellular disruption (Iqbal *et al.*, 1996). Inside the intracellular spaces O_3 will convert into other ROS including: hydroxyl radical, superoxide anion radical, hydrogen peroxide, and others. These ROS can lead to ultrastructural damage and can also oxidize the proteins of the cell membrane, called oxidative stress (Iqbal *et al.*, 1996).

Once exposed to O_3 the plant's defense system is triggered and several reactions can occur. Some higher plants have a natural defense system, which can eliminate the harmful effects of O_3 exposure via enzymes, antioxidants and radical scavenging compounds (Iqbal *et al.*, 1996). If the defense system is sufficiently strong, ozone-induced damage may be largely avoided. This occurs only in "ozone tolerant" plants (Iqbal *et al.*, 1996; Pell *et al.*, 1997). Plants such as milkweed cannot detoxify O_3 and ROS cause oxidative stress; these plants are considered "ozone sensitive" (Iqbal *et al.*, 1996; Pell *et al.*, 1997). Sensitive plants can respond to O_3 using mechanisms that result in cellular and metabolic changes, showing visible and often quantifiable injury (Pell *et* *al.*, 1997; Schraudner *et al.*, 1997; Rao *et al.*, 2000). It is for these reasons that some higher plants are useful for indicating the effects of ground-level O_3. Since the AQI and is a mechanical measures and do not take into consideration the cumulative or biological effects of O_3 exposure monitoring with plants can provide more biologically relevant information than a simple measurement.

3.3 Acute versus Chronic Ozone Exposure

Plants can react differently to short term and long term O_3 exposure (Tingey & Taylor, 1982). Acute exposure occurs when local O_3 levels are elevated (>100 ppb) for a few hours only (Rao *et al.*, 2000). This short burst of O_3 will produce immediate plant reactions and can result in visible lesions (Rao *et al.*, 2000). Acute exposure can also induce programmed cell death to limit the mobilization of a pathogen or pollutant. This is an example of the avoidance response known as compartmentalization, where the pollution filled compartment is isolated to prevent further cellular damage (Pell *et al.*, 1997; Schraudner *et al.*, 1997; Rao *et al.*, 2000). Short bursts of ozone can also cause cell death because O_3 decreases turgor pressure, resulting in deformation of the cell wall to the point of collapse (Pell *et al.*, 1997). Both of these responses are extreme and detrimental to the plant (Pell *et al.*, 1997).

Chronic exposure occurs when O_3 levels are elevated above 65 ppb, but less than 100 ppb for several days or weeks. The plant response does not initiate programmed or unregulated cell death and immediate injury formation does not occur, however, overall plant productivity and competitive ability is decreased (Pell *et al.*, 1997; Rao *et al.*, 2000). For the duration of the chronic exposure the cell membrane is thought to remain intact and the plant will continually respond to the pollutant (Pell *et al.*, 1997). Under these conditions the tolerance response is more effective at protecting against O_3 exposure and is less harmful to the plant than programmed cell death. However, as the plant naturally ages, defensive antioxidant levels will decrease and the chronic exposure can lead to premature senescence (Iqbal *et al.*, 1996; Pell *et al.*, 1997).

3.4 Types of Visible Ozone-Induced Damage

Over the course of the growing season plants can naturally be exposed to chronic and acute levels of O_3 and because of this can exhibit several types of visible ozone-induced damage. This visible damage can include chlorosis where there is reduced chlorophyll and the leaves appear yellowed or bleached; necrotic lesions, which are groups of dead cells; and foliar stipple, which appears as small black dots found between veinlets on the upper leaf surface (Appendix II). Stipples are made up of individual dead cells thought to be the combination of chlorophyll reduction and a pigmented byproduct, (Figure 3.1) (Kohut, 2005). Plants have also been known to have premature leaf drop known as accelerated foliar senescence. Premature leaf drop is difficult to quantify *in situ* and is not a reliable indicator of O_3 presence by itself, because senescence can occur for many reasons, including lack of moisture or nutrients. The types of ozone-induced damage are not always induced by acute vs. chronic exposures exclusively, when assessing plants *in situ* often times both regimes will occur in a growing season.

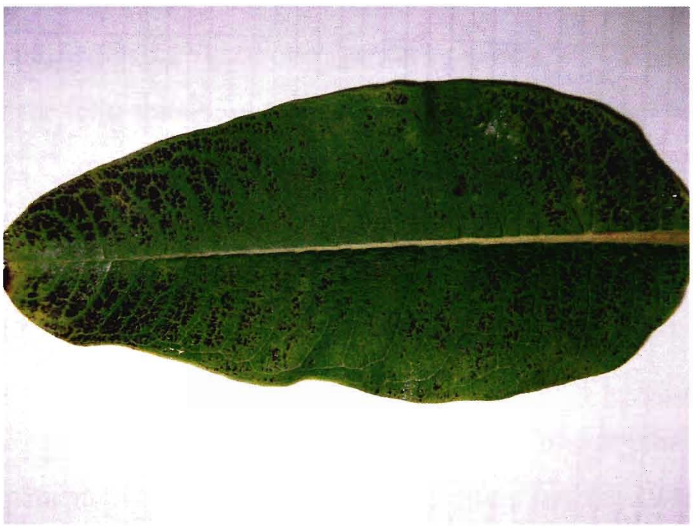


Figure 3.1: Image of a milkweed leaf (approximately 12 cm) with stipple (black dots) on the upper surface and interveinally due to cellular damage.

Although all of these symptoms are linked to O_3 exposure most of the injuries are difficult to visually assess accurately for the purpose of monitoring (Skelly, 2000; Kohut, 2005). The percentage of leaf area covered by chlorosis can not only be difficult to quantify, but can also lead to premature leaf drop, which may lead an investigator to possibly underestimate O_3 exposure due to the lack of leaves. In addition chlorosis and premature senescence are difficult to distinguish *in situ* from other stressors such as nutrient deficiency, drought and pest infestations (Kohut, 2005). For this reason foliar stipple is the most easily recognizable and visually quantifiable ozone-induced damage making it the most widely used visual indicator (Skelly, 2000; Kohut, 2005; Davis & Orendovici, 2006).

3.5 Identifying Ozone-Induced Damage by Adaxial Stippling

Stippling on the upper leaf surface is the most easily recognizable form of ozone-induced injury (Skelly, 2000). The stipples are individual dead palisade and spongy mesophyll cells found on the upper leaf surface that appear as black dots. Stippling is distributed on the vascular tissue (veins), not the stomata (Faoro & Iritti, 2005).

Ozone-induced injury that is used in biomonitoring with milkweed plants has long been quantified using visual observation by presence or absence of stipple or estimation of the total amount of leaf surface area that is covered with black dots (Kohut, 2005). Ozone-induced injuries have been classified in studies using grape plants as indicators in California, agricultural plants as indicators in Ontario, and milkweed in Great Smoky Mountains National Park, Shenandoah National Park, and National Wildlife Refuges (Richards, Middleton and Hewitt, 1958; Pearson, 1992; Chappelka *et al.*, 1997; Davis & Orendovici, 2006; Neufeld *et al.*, 2006). Such data can then be quantified using the Horsfall-Barratt scale, a common plant injury classification system with percent of injury categories (Horsfall and Barratt, 1945). In recent years some researchers in the ozone monitoring community have adopted a modified Horsfall-Barratt scale, which has six injury classes instead of the original 12 as it is said to be more suitable for field work (Chappelka *et al.*, 1997; Skelly, 2000). The percent cover classes are: 0 = 0% coverage, 1 = 1-6%, 2 = 7-25%, 3 = 26-50%, 4 = 51-75%, 5 =

76-100% (Chappelka *et al.*, 1997; Skelly, 2000). Because stippling is an irreversible injury the amount of coverage increases through the ozone season. For this reason at the end of the O_3 season, prior to leaf drop the amount of stippling on an affected leaf should reflect the exposure for the whole season.

3.6 Classic Studies of Ozone Monitoring using Milkweed Plants

Ozone-induced injury to plants was first reported by unnamed citizens in California in 1944 (Griffiths, 2003). Since O_3 causes a characteristic and quantifiable black-purple stippling on grape leaves (Richards *et al.*,1958) there have been dozens of studies using grape plants as indicators of ozone presence by government workers and university researchers. Ozone monitoring studies have been reviewed by Duchelle & Skelly, 1981; Chappelka *et al*, 1997; Pell *et al.*, 1999; Gunthardt-Goerg *et al.*, 2000; Kouterick *et al.*, 2000; Skelly *et al.*, 2000; Lee, Steiner, Zhang & Skelly, 2002; Yuska *et al.*, 2003; Davis & Orendovici, 2006; Souza *et al.*, 2006. The following section will give a review of these historical studies.

It is known that plants are sensitive to ozone as shown by the early studies beginning with Richards *et al.* in 1948. One such pioneering, controlled chamber study conducted by Duchelle & Skelly (1981) sought to determine if common milkweed plants were sensitive to O_3 exposure and if so to determine the thresholds at which ozone-induced damage were apparent. The study had both field and laboratory components. In the laboratory, milkweed were fumigated at 11 and 26 weeks of age (after germination) for seven days with one of four concentrations of O_3 (0, 50, 100, 150 ppb 6 hr-/day). In the field experiments conducted in Virginia, three treatments were applied to 3.0 m diameter chambers where naturally growing milkweed would emerge. One group of plants received ambient air in an open-top chamber, while another received charcoal-filtered air and the last received unfiltered air via fans in a closed chamber. The milkweed plants were monitored for the presence of insufficient chlorophyll production (chlorosis) and stipples. It was found that the milkweed grown in the presence of O_3 (grown in ambient air and grown in unfiltered air) showed signs of chlorosis and stipples by the middle of June. The plants grown in charcoal-filtered air had no ozone-induced damage. Both 11 and 26-wk old plants grown in the lab subjected to zero O_3 exposure did not show any damage, the 11-wk old plants fumigated with 50 ppb had 62% of the plants showing ozone-induced stipples, and 26-wk old plants had 6% exhibiting stipples. Under the 150 ppb exposure 100% of the plants showed ozone-induced stippling. The authors determined that because the 11-wk old plants had a higher incidence of damage, plant age at time of exposure was an important factor. This was one of the first studies to show milkweed response to O_3 exposure. It concluded that milkweed plants were sensitive to O_3 and that ozone-induced damage was quantifiable. The finding of high variability in injury between plants that received the same exposure to O_3 suggested that large sampling sizes may be necessary when biomonitoring with milkweed. Because this was an early study the exposures were quite high, however, today we know that fumigating plants with 150 ppb of O_3 is excessive. A value of 150 ppb is rarely seen in ambient environments and is therefore unrealistic. These findings would influence future research because it was the first to show that milkweed could be used as biomonitors.

After the first studies using milkweed in fumigation chambers came the non-chamber studies using milkweed indicators growing in natural conditions. In these studies milkweed were assessed for foliar damage and compared to known levels of O_3 exposure. One of the first studies of ambient ozone-induced damage to milkweed plants *in situ* without the use of chambers was completed by Chappelka *et al.* (1997) in the Great Smoky Mountain National Park. Milkweed plants were assessed along pathways (not plots) in the park for damage twice during the growing season on August 03-08 and August 25-28 (1992) using the modified Horsfall-Barratt scale. Milkweed growing in ditches were not chosen because they would not be representative of the moisture regime of the area due to water accumulation. This factor is important because moisture availability effects stomatal opening, which in turn effects O_3 uptake. Also, sites next to a road could have confounding effects on normal plant growth, including variations in other pollutants associated with automobiles, and road salt residuals. The data were collected by teams trained in assessing ozone-induced damage to all milkweed plants (N = 1300) in the study area. The information was reported as the foliage injured (% of leaves injured) and leaf area injured (%), which was then categorized into Horsfall-Barratt classes (0-5). Severity averages were determined by taking the mid-point of the

Horsfall-Barratt class. Ozone exposure data were obtained from three air quality stations in the park. In early August, 74% of the milkweed plants exhibited O_3 damage and by late August 79% of the plants were damaged. This was the first study to show the large scale ambient ozone-induced damage to milkweed plants in the natural habitat. It would be interesting to have known more information regarding the milkweed stands and microenvironmental conditions that were observed. However, this methodology of surveying milkweed for percent of plants and leaves damaged would serve as a model again in future research.

Davis & Orendovici (2006) conducted a multiyear study in a National Wildlife Refuge (New Jersey) where several plant and tree species were assessed to see if they exhibited ozone-induced sensitivity and damage. The purpose was to define the exposure-damage relationships of these potential bio-indicators using the most useful O_3 index, and to assess what role soil moisture levels played in inhibiting damage. Ozone damage is cumulative in higher plants and therefore injury can occur rapidly during the final month of the O_3 season. The surveys were conducted at the end of the season in August for several years (1993-1996, 2001-2003) and were qualitative in nature reporting damage as presence or absence of stippling only. The study assessed seven sensitive species, which included common milkweed, wild grape, tree-of-heaven, Virginia creeper, winged sumac, black cherry, and sassafras at two regions in the refuge. At ten areas within each region a minimum of 50 plants of all seven species were surveyed (N = 500/species) and the number of plants with ozone-induced damage/total plants per species was calculated and defined as "incidence". The incidence was compared to known O_3 levels as determined by the one permanent monitoring station in the refuge. The O_3 data were reported as SUM0, SUM60, SUM80 indices, which add values of O_3 over a certain level (e.g., SUM0, SUM60, SUM80 are all values summed over 0, 60, and 80 ppb, respectively). The other O_3 reporting metrics were the W126, which assigns larger values to larger O_3 exposures, the N100, which sums the number of hours the O_3 breaches 100 ppb, and the 12-hr and 24-hr average. Based on previous studies the authors chose to compare SUM60 to ozone-induced damage to show the correlation. Since plant damage is in part a function of turgor pressure and water availability, many researchers use a drought index to infer water availability. It was thought that an indication of soil moisture was needed and the Palmer Drought Severity Index (PDSI) was used which gives a value based on the soil moisture deviations from normal. Although an important step was made in assessing the moisture regime of the area, the PDSI is a generalized index that determines a relative drought value based on historical levels and commonly assigns one value for a very large region. This methodology might underestimate fine-scale differences in precipitation locally. Additionally, the PDSI is a regional index and not a direct measure of soil moisture nor turgor pressure in the plants. It is not expected to provide a close correlation if there are microclimatic differences.

Mathematical models using binomial logistic regression were used to establish if there was a correlation between the incidence of plant damage and O_3 indices. These were tested to characterize ozone-induced damage by identifying the most suitable O_3 index and which factors of the plant environment were related to plant damage. The PDSI for soil moisture gave the best fit of the indices that were explored (12-hr average, 24-hr average, SUM0, SUM60, SUM80, W126, N100). The model with the best fit utilized both the W126 and N100 along with the drought index to predict O_3 damage. Linear regressions were used to determine that common milkweed was the most reliable indicator to consistently show ozone-induced injury. Species that were not considered to be “sensitive” (Virginia creeper, singed sumac, black cherry, sassafras) either did not show consistent incidence of damage or the damage was not attributed to O_3 exposure alone, but to a combination of nutrient stress, insect damage.

The Davis & Orendovici study has two significant findings for ozone monitoring with milkweed. First, O_3 exposure data alone were not sufficient to characterize O_3 damage to plants, it was necessary to input a parameter, which accounts for soil moisture. Second, the mathematical models showed that the use of one index did not accurately characterize biologically relevant O_3 exposure. The model using a seasonally cumulative ozone index (SUM) with the drought index showed a moderately good fit for milkweed. However, the weighted indices (W126, N100) when used together give a more biologically relevant indication of the O_3 exposure that will induce plant damage at this location. This is true because the W126 and N100 indices are weighted for higher O_3 exposure, which is useful because the highly elevated exposures are important for inducing plant damage, but can miss total ozone data that is accounted for in cumulative indices.

Another multiyear study that relates milkweed O_3 damage to the corresponding O_3 indices was conducted by Bennett, Jepsen & Roth (2006). In it, a total of 18 permanent plots were established on either side of Lake Michigan in order to compare the different regimes of O_3, soils and climate, and to characterize how these factors will effect ozone-induced damage. In that work, O_3 sensitive black cherry trees and milkweed plants were continuously surveyed over three years in their natural growing conditions while exposed to ambient O_3. Ozone exposure data were obtained from the US EPA and climatic data were obtained from 39 climate stations. Quantitative assessments were made for plant health on 78-150 plants per site (N = 1404-2700). This included ozone-induced damage using the modified Horsfall-Barratt scale, insect and other disease damage, and soil collections taken at 0-20 cm depth. The soil samples were analyzed for texture and elemental, organic, and nutrient content in order to characterize the growing conditions below ground. The data obtained for air temperature and precipitation for each study year were assessed as departures from climatic normals in order to characterize climate. Ozone exposure data retrieved from the US EPA was in the form of SUM00, SUM06 (ppm), W126, and AOT40 indices (cumulative index: Europe). The first objective was to determine the relationship between O_3 exposure, soil texture and climate for each species. Secondly, the authors were interested in O_3 effect on plant height and seed pod number.

Through classification and regression tree (CART) analysis it was determined that the SUM00, SUM06, W126, and AOT40 indices were not correlated to O_3 exposure to plant damage (Bennett *et al.*, 2006). Similar to Davis and Orendovici's findings (2006) each index when assessed independently did accurately define the actual O_3 exposure, but were not representative of the relationship between O_3 exposure and plant damage. It was determined that the W126 when used with the N100 was closely correlated to ozone-induced damage. The combination of the two indices is useful because the W126 was created based on biological relevance where exposures over 40 ppb are weighted more heavily, while the N100 takes into account the periodic highly elevated O_3 levels that tend to occur in ambient air.

Bennett *et al.* (2006) found that O_3 damage to milkweed plants was more consistent than on black cherry. The relationship between ozone-induced damage and O_3 exposure was significant with a p-value of 0.05. Soil chemistry, rainfall and O_3 exposure had a significant effect on milkweed height at a p-value of 0.05. In the case of milkweed the seed pod number showed a significant inverse relationship to O_3 exposure. The authors suggest that O_3 exposure may have effects on the reproductive potential as measured by seed pod number, however, the impact of O_3 exposure on future milkweed populations was not addressed in the Bennett *et al.* (2006) study and further investigation would be necessary. As with all the previously mentioned studies the results show that *in situ* ozone biomonitoring is possible when the effect of the microclimatic factors are understood.

In these studies O_3 concentrations were obtained from government regulated air quality sensors. Portable sensors can also be used to record continuous O_3 concentrations for up to one week as a means of correlating damage to exposure. This methodology is useful if there is a lack of permanent sensors or access to the data is limited. In a study conducted in north central Pennsylvania by Yuska *et al.* (2003) portable sensors were compared with continuous sensors when assessing damage to the O_3 sensitive black cherry trees and milkweed plants. Damage was visually assessed weekly on the same 15 milkweed plants and 15 black cherry trees using the modified Horsfall-Barratt scale at 15 sites (N = 225/species) from May-September for two years. Average weekly O_3 exposure data were obtained from portable O_3 sensors at each site, the filters for which were extracted on a weekly basis to retrieve the data, and from continuous O_3 analyzers for comparison. The O_3 data were reported as the cumulative seasonal concentrations (ppb), and seasonal O_3 averages (ppb) per site. Available soil moisture was determined weekly using soil moisture blocks attached to a moisture tester at 10-12 cm depth. The authors attempted to determine if ozone-induced damage is related to O_3 exposures, and to identify the point at which O_3 concentrations would induce damage when moisture is taken into account.

It was found that the portable sensors were able to reliably determine O_3 concentrations evidenced by strong correlations to the continuous sensor data showing that portable and continuous sensors could be used interchangeably. Milkweed plants showed ozone-induced damage at all study sites by September, but also exhibited more severity when compared to black cherry. It was found that milkweed O_3 damage was correlated to the cumulative seasonal O_3 concentrations with an r-value of 0.658 as determined by correlation and regression analysis. Because that study was conducted at several different sites with varying amounts of precipitation the soil moisture data were an important factor in understanding the distribution of milkweed damage. It was determined that the site with highest seasonal rainfall (55.63 cm) showed the most milkweed damage of all the sites. Similarly, sites with low moisture reflected low damage regardless of elevated O_3 levels. Wilted leaves were not assessed in that study. The injury to black cherry trees was found to be significant determined by the relationship between cumulative O_3 and soil moisture (p-value =0.001). The authors determined that this was due in part to the optimal conditions for plant growth, which will promote O_3 uptake. This trend was present and observed for milkweed plants, but was not significant, which could have been due to the variability in soil moisture, or plant injury. In that study actual measurements of soil moisture were taken as opposed to using an index. However, the measurements were captured only once weekly and could account for the insignificance in the milkweed test. Soil moisture data have potential, but the variability may be large and may go undetected if sampling is done infrequently. However, in the absence of plant moisture data measuring soil moisture may be a crude, yet effective way of determining the moisture status of the plants' surroundings.

Bergweiler *et al.* (2008) chose to model the exact net photosynthesis and stomatal conductance of the milkweed leaves to determine O_3 uptake for two growing seasons. Milkweed plants were grown in an open field with two treatments: one with added water and the other without. Throughout the growing season measurements for air temperature, relative humidity, photosynthetically active radiation (PAR), wind direction and velocity, and soil water content were recorded using data loggers. Each leaf set (pair of leaves) was marked and measured for net photosynthesis and stomatal conductance daily. It was found that stomatal conductance and therefore O_3 uptake was different based on the water treatment, and that increased uptake reflected an increased moisture regime. The authors determined that during the dryer year the site with water added had higher O_3 uptake. However, during the year with increased natural precipitation the stomatal conductance differences were less pronounced. Stomatal conductance differences were also reduced toward the end of the growing season as the plants aged. This indicates the importance of O_3 exposure timing during the growing season, as younger plants seem to have higher gas exchange. The authors indicate that this is one of the first studies to closely examine the physiological effects of microenvironment on ozone-induced damage to plants. It is suggested that the effects are species-specific and that moisture regime as it affects O_3 uptake is an important factor in producing O_3 damage. They go on to further explain that this is an emerging field and a complete characterization of the plant-atmosphere interactions will be important to the continued use of what is described as an "important bio-indicator species". The Bergweiler *et al.* (2008) study highlights the importance of characterizing moisture regime when using milkweed as indicators. The researchers show that with accessibility to instrumentation plant moisture can be accurately determined.

Some studies have quantified factors such as nutrient deficiency and insect damage, which could possibly add to the stress of the plant. However, to definitively say that a plant was stressed would necessitate accurate data collection on the nutrient status of the soil and a knowledge of insect identification and damage.

In each of the *in situ* O_3 monitoring studies using milkweed the methodologies for assessing damage are similar and there remain three important factors. This includes monitoring actual O_3 exposure using either continuous or portable samplers, determination of O_3 damage either quantitatively or qualitatively, and quantifying soil moisture in order to characterize the growing conditions of the plant. It is important to understand that not all O_3 exposure indices are biologically relevant, some indices simply report on the levels of O_3, but not all O_3 levels will cause ozone-induced damage. Also the microclimatic variations at each site, where soil moisture appears to be the most important, will result in differing levels of visible O_3 plant damage. Some of these studies simply extrapolated the moisture status of the plants by obtaining the drought indices or precipitation data for the whole study region (Yuska *et al.*, 2003; Davis and Orendovici, 2006). Finally it is important to remember that biomonitoring will never replace permanent sensors, but can be used for preliminary assessments that allow for a cost-effective canvassing of a region so that areas of interest may be discovered and further research can be completed. Biomonitoring studies using agricultural crops have been conducted in Ontario for decades, however, I was unable to find any published studies that have used milkweed as a bio-indicator of ozone exposure in Ontario (MOE, 1989).

Chapter 4: Study Design and Methods

4.1 Study Sites

AQI stations were identified within southern Ontario and milkweed sites were established as close to the AQI stations as possible. All sites were within five kilometers of the stations, (Figure 4.1). This allowed ozone-induced milkweed damage to be compared to the O_3 exposures recorded, (Table 4.1, 4.2). Eight study sites (Essex, Sarnia, Chatham, Grand Bend, London, Port Stanley, Simcoe and Brantford) were selected using the following criteria: the milkweed stands had at least 100 plants within 500 meters of each other, and were not found in drainage ditches nor within 100 meters of a highway. All of the study sites were open grown, on public land except for the Simcoe site, which was owned by a local farmer who was contacted and permission was granted (Appendix I). Plants that exhibited mechanical damage or missing leaves because of animal presence were not included in this study. It should be noted that all of the sites had very little mechanical and insect damage.

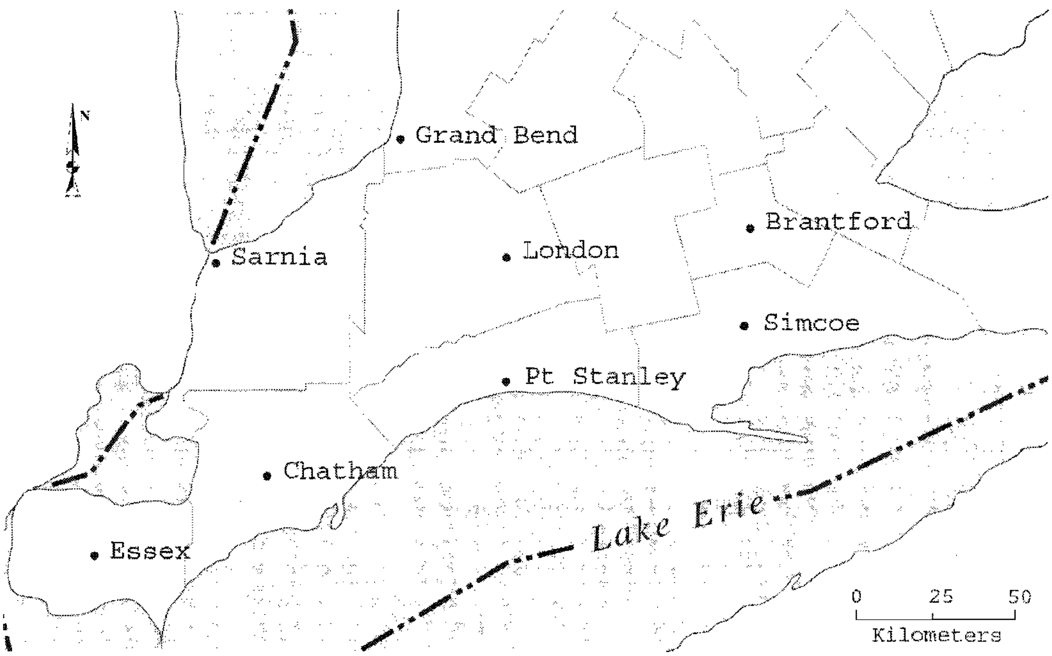


Figure 4.1: Location of eight study sites in southern Ontario (Map base supplied by Brock University Map Library).

Table 4.1: AQI stations where O_3 data sets were recorded, and meteorological stations (Met) where precipitation and temperature data were recorded. Survey sites were located in strips or patches along trails and walkways.

Site	Station ID	Latitude (degrees N)	Longitude (degrees W)	Type of Site	Site Size (square m)	Land Use	Soil	Distance from trees
ESSEX	AQI: 12059	42°09'36"	82°50'00"	Field	625	Residential	Clay	No trees or shrubs
	Met: 6133362	42°18'00"	82°54'00"	N/A				
CHATHAM	AQI: 13001	42°24'12"	82°12'31"	Park	300	Urbanized	Silt	5 m
	Met: 6131415	42°23'40"	82°13'20"	N/A				
SARNIA	AQI: 14064	42°59'01"	82°24'16"	Park	500	Urbanized	Silt	5 m
	Met: 6127519	43°00'00"	82°18'00"	N/A				
GRAND BEND	AQI: 15020	43°20'02"	81°44'20"	Trail	400	Residential	Sand	10 m
	Met: 6122370	43°21'00"	81°30'00"	N/A				
LONDON	AQI: 15025	43°00'32"	81°12'34"	Park	225	Residential	Sand	5 m
	Met: 6144478	43°18'00"	81°09'00"	N/A				
PORT STANLEY	AQI: 16015	42°39'36"	81°13'08"	Field	500	Agricultural	Silt	10 m from crops, 15 m from trees
	Met: 6137362	42°46'20"	81°12'60"	N/A				
BRANTFORD	AQI: 21005	43°08'25"	80°17'46"	Park	130	Residential	Clay	15 m
	Met: 6153194	43°10'00"	79°55'80"	N/A				
SIMCOE	AQI: 22071	42°51'08"	80°15'50"	Field	750	Agricultural	Clay	No trees or shrubs
	Met: 6137154	42°27'00"	81°52.80'	N/A				

Table 4.2: Data collected from 75 milkweed plants at eight sites in Ontario, O_3 indices calculated from raw data received from the Ministry of the Environment for the O_3 season June-September, 2007. Authors who implemented the methods or measurements are found in "Reference". (Modified Horsfall-Barratt Scale = H-B).

Measurement	**Methods**	**Number of Measurements**	**Units**	**Reference**
Plant height	Yard stick	1 per plant	± 1 cm	
Number of leaves	Count	1 per plant	number	
Number of O_3 damaged leaves	assessed for stipple 1 leaf = 1 count	all leaves on plant	number	Chappelka *et al.*, 1997; Skelly, 2000; Yuska *et al.*, 2003; Kohut, 2005
Area of injured leaf	visually assess the area of stipple (percent cover)	all leaves on plant	H-B	Chappelka *et al.*, 1997; Skelly, 2000; Yuska *et al.*, 2003; Souza *et al.*, 2006
Soil Texture	soil extracted at ~25 cm (~25 g), texture assessed	1 per site	Soil type	Bennett *et al.*, 2006; Davis & Orendovici, 2006
Soil moisture	soil extracted at ~25 cm measured wet weight, oven dried weight	1 per site	±1 milligram	Bennett *et al.*, 2006; Davis & Orendovici, 2006
Calculations				
Incidence (leaves damaged)	damaged leaves/all leaves	1 per plant	%	Chappelka *et al.*, 1997; Davis & Orendovici, 2006; Souza *et al.*, 2006
Incidence (plants damaged)	number of damaged plants/75	1 per site	%	Chappelka *et al.*, 1997; Davis & Orendovici, 2006; Souza *et al.*, 2006
Average H-B Value	sum of Horsfall-Barratt scale/number of leaves	1 per plant	H-B	Chappelka *et al.*, 1997; Skelly, 2000; Yuska *et al.*, 2003; Souza *et al.*, 2006
Total O_3	Incidence-plants x average Horsfall-Barratt value per plant	1 per plant	H-B	Bennett *et al.*, 2006
Soil moisture	(wet weight-dry weight)/dry weight	1 per site	%	Bennett *et al.*, 2006
Ozone Indices				
SUM65	sum each hour over 65 ppb	24 hours/day	ppb	Chappelka *et al.*, 1997; Davis & Orendovici, 2006; Souza *et al.*, 2006
SUM80	sum each hour over 80 ppb	24 hours/day	ppb	Chappelka *et al.*, 1997; Davis & Orendovici, 2006; Souza *et al.*, 2006
Cumulative O_3	sum all O_3 values over zero	24 hours/day	ppb	Davis & Orendovici, 2006
1-hour >65	sum all hours over 65 ppb	24 hours/day	hours	Bennett *et al.*, 2006; Davis & Orendovici, 2006
1-hour >80	sum all hours over 80 ppb	24 hours/day	hours	Bennett *et al.*, 2006; Davis & Orendovici, 2006
12-hr average	ppb average of 0800-2000	24 hours/day	ppb	Davis & Orendovici, 2006
24-hr average	total ppb 24-hour average	24 hours/day	ppb	Davis & Orendovici, 2006

4.2 Field Methods and Data Collection

The methodology used here closely resembles that used by Yuska *et al.* (2003), Bennett *et al.* (2006), and Davis & Orendovici (2006). This study differs from the others as it was conducted over one field season, and only milkweed plants were used. The advantage of using only milkweed plants is that several sites were found which may not have been possible if several species were used. Also, with the constraints of only one researcher conducting the surveys one species was more feasible. The same plants were not tagged and inspected at each survey due to possible loss of study plants because the study sites were not protected. However, the same plant stands were visited at each survey, which reduced microclimatic variation. Milkweed plants that were dead, visibly damaged from insects or broken where not surveyed. At each site there were few overall damaged plants that were omitted so this was not considered to be an important factor as there were plenty of other plants to survey. Measurements included plant height where a wooden yard stick was placed within 10 cm of the plant base and held perpendicular to the ground, the measurement was read to the nearest centimeter. Stem number, and leaf number were counted per plant. In addition, foliar injury defined by adaxial (upper leaf surface) stipple was visually determined by the author and assigned Horsfall-Barratt value per leaf, (Table 4.2). Images of typical milkweed injury can be found in Appendix II.

The eight station sites were located in early-May to determine what environment the AQI stations were located in, (e.g., industrial area, park, trail, etc.), if milkweed plants could grow nearby, and if the sites fell within the pre-determined constraints (e.g., minimum number of plants, not in a ditch, nor on a roadway, etc.). Plant surveys began at the start of the growing season in late-May to determine if milkweed had emerged; the full assessment surveys were conducted in the middle of the growing season in June and in July and three times during the month of August (Aug 06-08; Aug 19-21; Aug 26-28) and into the first two weeks of September (Sept 04-05; Sept 10-11). Surveys were terminated at this time because the effects of regular senescence, including leaf drop, were not known and because the peak O_3 season historically, which runs from June to August had already passed. These factors could mask or introduce errors to the data. But, these potential sources of error were largely avoided by ending the surveys prior to leaf-drop in September. This was the most reasonable approach, one that is used in nearly all milkweed studies.

For each survey, 75 plants were inspected within the same site on the following dates: June 9-11, July 17-20, August 06-08, August 19-21, August 26-28, September 04-05, and September 10-11, (Table 4.2). The survey dates were chosen do determine exactly when the O_3 damage would occur and a concentrated weekly survey effort began in August. The number of plants chosen was based on the best practice found in the ozone-milkweed recent literature where plant numbers ranged from 15-150 per site. Since the sites were in urban environments it was difficult to find plots with over 200 plants. This decision was also based on time constraints, as 75 plants took up to two hours, 150 plants could have taken twice as long and was not logistically feasible. Additionally the milkweed variability at each site seemed to be low so 75 plants was determined to be sufficient. Each survey involved approximately 900 kilometers of driving over three days. It was decided to keep the surveys limited to three days because during August and September the surveys were to be completed weekly and a 5-6 day survey was not possible. The timeframe was adequate for surveying 75 plants per site. Many of the sites had milkweed stands with approximately 100-150 plants, but one site (Essex) had approximately 300 closely grouped plants.

In previous studies a region-wide meteorological index was used to characterize the precipitation for the study area. However, in this study soil moisture and texture were determined for each site. One soil sample (20-30 g) was removed at milkweed rooting depth (20-25 cm) at or near the center of each survey site. Sampling was done at each visit. The samples were placed in aluminum containers wrapped with rubberized tape to prevent moisture loss. The soil was transported to the lab and analyzed within five days. The wet weight of each soil sample was weighed carefully to ±0.0001 g accuracy on a Sartorius balance (Type 1702, Göttingen, Germany). The samples were then placed in a 100°C oven for 24 hours to remove the moisture and weighed again with the same analytical balance to determine the dry weights. From this the percentage of water (% water) was calculated. This approach was believed to give an accurate measure of the wet versus dry weight of the soil samples. It was thought that due to differences in precipitation across the study region collecting soil and calculating %

water would give a more accurate indication of the moisture status of the plants. Total daily precipitation and mean daily air temperature values were also retrieved from Environment Canada Climate Weather Office (Environment Canada Climate Weather Office).

4.3 Calculations

The calculations for SUM65, SUM80, hours > 65 ppb, hours > 80 ppb, N100, 12-hr average, 24-hr average, and seasonal cumulative O_3 derived from the raw and collected data can be found in (Table 4.2). The average Horsfall-Barratt values for each plant were calculated from the mid-point of each class resulting in an average Horsfall-Barratt class value per plant (Chappelka *et al.*, 1997). Total O_3 injury was calculated by taking the actual precedence of O_3 injury per plant (incidence) and multiplying it by the Horsfall-Barratt value resulting in a number on a 0-5 scale. The peak 1-hr value per day was also determined for the site that had the highest O_3 each day.

4.4 Data Analysis

Unpublished O_3 data were obtained for the eight AQI monitoring stations from Mrs. Laura Fiore of the Terrestrial Assessment Unit Air Monitoring and Reporting Section of the Ontario Ministry of the Environment (MOE). The data were provided under a data sharing agreement signed by Professor D. McCarthy of the Department of Earth Sciences at Brock University and the MOE. These data were made available in raw, unverified form at least 6 months prior to their publication in the AQI report. The hourly data were obtained in parts per billion (ppb) as Microsoft™ Excel files that covered the dates June 01-September 30, 2007 and the indices most commonly reported by the O_3-milkweed monitoring community were calculated. There was a total of 23, 328 values of O_3 hourly data (ppb) for the study period. There were minimal gaps in the records, approximately 75 that were dispersed throughout the whole season. These were counted as zeros. This was done in order to avoid adding false values to the data, and because the ratio of missing data was so small it did not significantly change the outcome. Port Stanley had two days (48) of missing data, which was the highest, followed by Essex with 17 missing data points, Grand Bend with 3, and Sarnia with 1.

Inserting a zero in a blank record was a conservative approach that underestimated the true SUM65 and SUM80. However, even with two days of missing data Port Stanley's missing measurements represent only 0.0019% of the data set. The SUM indices calculated were the SUM65 and SUM80 (ppb), cumulative O_3, 1-hour exceedances (> 65 and 80 ppb), 12-hr average and 24-hr average. The W126 index which is used in reporting ozone-induced damage is too complex a mathematical calculation to be used in this study due to the time it would take to complete the calculation for all sites.

Ozone exposure indices were calculated from the raw data to determine which one would best characterize the O_3 regime of 2007. Total daily precipitation and mean daily air temperature values were analyzed to determine the link between O_3 exposure, plant injury and meteorological conditions. The total daily precipitation was a more complete set of data (one value per day) versus the soil moisture measurements that were taken per survey. The O_3 exposure indices, O_3 milkweed damage, total daily precipitation and mean daily air temperature data were assessed statistically using the mixed procedure, univariate analysis of variance (ANOVA), and multivariate analysis of variance (MANOVA) with repeated measures using SAS statistical software, version 9.1 for windows (SAS Institute Inc., Cary, NC).

Ozone-induced damage was reported as four different dependent variables so that the most accurate and efficient way of quantifying O_3 damage could be determined by statistical analysis. The incidence of plants damaged per site was calculated, as was the incidence of leaves damaged per plant, the average Horsfall-Barratt value per plant and the total O_3 per site, (Table 4.2). It was decided to focus on three dependent variables for characterizing O_3 damage because they were the most commonly reported. These were the incidence of leaves damaged per plant, incidence of plants damaged per site and total O_3 damage. The average of the 75 observations was used to simplify the analysis and modeling. This decision was based on normal statistical practice of using either an average or median.

The dependent variables were assessed statistically using ANOVA and MANOVA to determine which one was most useful in characterizing milkweed O_3 exposure. The surveys that had evidence of O_3 damage were subjected to statistical analysis, for each survey there were 3000 data points. For all statistical analysis an *F*-

value less than 0.05 was considered to be statistically significant. The data entered into SAS 9.1 can be found in Appendix III, SAS outputs in Appendix IV, the table for *F* critical values in Appendix V, and raw data in Appendix VI.

The mixed procedure was utilized for the ANOVA analysis and the assumptions of constant variance and normality were made and met by producing and visually assessing the residual plot for linear distribution and qq plot for normal distribution, respectively. The General Linear Models (GLM) procedure was utilized for the MANOVA analysis and the assumption of normality was met by assessing the qq plot. The sites were visited several times, so the repeated measures model was used to relate each survey per site.

The timing of the surveys (time 1 = survey 1, time 2 = survey 2, etc.) was used as a factor because the O_3 damage is changing across the study period. For the MANOVA analysis the repeated measures model was used. Repeated measures is used when a parameter is assigned a value more than once during the analysis and the values are not independent of each other. It was necessary to use repeated measures to avoid a falsely rejected null hypothesis based on the fact that the same site was visited repeatedly and each survey was not independent of the previous survey results. To check the assumptions made by using the mixed model with repeated measures normality and constant variance were assessed and found to be random, indicating there was no pattern. Temporal pseudoreplication occurs when replicates are not observed in one survey, but are instead accumulated at several surveys across time. Finally, the between site values were assessed independently. No assessment was made of within site variability and the assumption is that it was negligible.

Note: "The *F* test determines if the two values being compared aren't different. Small differences (and *F* values not very different from 1.0) might come up often if the variances were the same, but that big differences (and *F* values that are quite different from 1.0) would come up less commonly. We use these probabilities to decide if our results would happen very often, if the real variances were equal. If our calculated *F* ratio turned out to be uncommonly large, we could make the decision that maybe the variances really aren't equal. In many areas of biology the 5% level of probability is used because if the *F* ratio we calculated would happen only 5% of the time if the variances really were equal, we could and proceed with the assumption that the variances really are different, and accept a 5% chance of being wrong." (SFSU, 2009). The F value is compared to the *F* critical table (Appendix V), if the *F* value exceeds the F critical the result is statistically significant. The findings are reported in the following format: F (degrees of freedom (numerator), degrees of freedom (denominator))= "F-value", p= "p-value".

Chapter 5: Results

5.1 Ozone Exposure and Indices

Table 5.1 shows the nine O_3 indices that were calculated, including the number of one hour exceedances above 65 and 80 ppb (hr>65 and hr>80), the number of hours above 100 ppb (N100), the 12-hr average (12-hr), which is the average O_3 (ppb) between the hours of 800-2000, the seasonal cumulative O_3 (Cum O_3) (ppb), and SUM65 and SUM80 are the additive ppb above 65 and 80, respectively.

Port Stanley had the highest 12-hr average O_3 (45.56), followed by Simcoe (44.73) and Chatham (44.05). The highest 24-hr averages were located at Simcoe (38.05), Pt. Stanley (38.44) and Chatham (37.39). Simcoe (112,234) reported the highest values for Cumulative O_3 with Chatham (109,376) and Pt. Stanley (108,862) showing the second and third highest, respectively.

Port Stanley had the highest number of hours above 65 ppb (232) and above 80 ppb (66), (Table 5.1). In total, Port Stanley spent 232 hours in non-attainment of these standards and the second closest was Chatham with 185 hours above 65 ppb, (Figure 5.1). Grand Bend had 10 hours in excess of 100 ppb. The highest peak 1-hr of 131 ppb was recorded on June 25 at Sarnia.

Using the peak 1-hr values it was determined which sites had the highest values each day, which represented elevated O_3 days. For the top three days of the 2007 season 24-hr backward trajectories were plotted to determine where the O_3 came from (Appendix VII). It was found using the NOAA website that on three elevated O_3 days the air parcels that ended in the study region originated in eastern Michigan, Ohio and the mid-western states before traveling across southern Ontario.

5.2 Ozone-Induced Damage and Indices

The incidence of leaves damaged at Essex at the end of the season (September 11) was 57.4%, 41.9% at Port Stanley and 37.5% at Simcoe, (Figure 5.2). The incidence of plants damaged per site showed that Essex had 89.33% plants damaged, Simcoe had 76%, and Port Stanley had 68% at the end of the O_3 season, (Table 5.2).

Table 5.1: Ozone exposure indices calculated from raw data received from the Ministry of the Environment for the O_3 season June-September, 2007. Calculations include: hours over 65, 80 ppb (hr>65, 80), hours over 100 ppb (N100), 12, 24-hour average (12-hr, 24-hr), cumulative O_3 (cum O_3), sum of concentration for hours over 65, 80 (SUM65, SUM80), number of days with the highest 1-hr O_3 for all sites (# days peak 1-hr). Bold values are the highest in each category; 0=zero.

Site	hr>65	hr>80	N100	12-hr	24-hr	CumO3 (ppb)	SUM65 (ppb)	SUM80 (ppb)	#days (peak 1-hr)
Pt. Stanley	**232**	**66**	1	**45.56**	38.05	108862	**18056**	**5827**	**51**
Simcoe	181	36	4	44.73	**38.44**	**112234**	13585	3125	18
Chatham	185	45	1	44.05	37.39	109376	13869	3870	15
Essex	150	26	0	42.35	34.18	99448	11113	2270	10
Gr.Bend	168	60	**10**	41.15	34.17	98341	13175	5437	17
Sarnia	133	42	3	40.05	33.37	96235	10242	3724	9
Brantford	156	16	0	42.34	32.55	95229	11490	1390	8
London	101	6	0	39.58	32.46	94930	7216	496	2

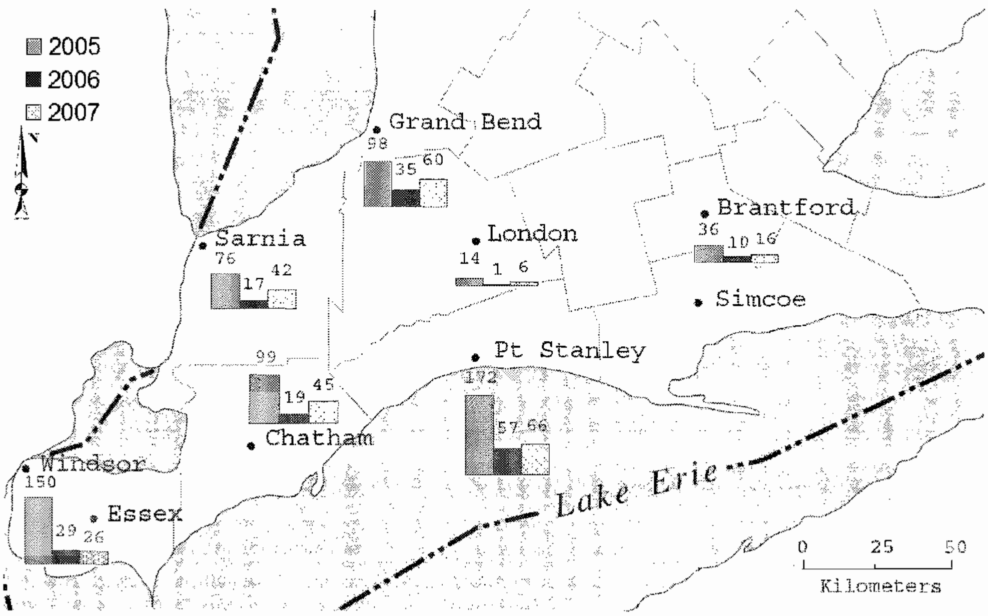


Figure 5.1: Locations of 8 study sites with data collected from 7 Air Quality Index stations (2005 and 2006), bars indicate the 1-hour exceedances > 80 ppb, which is the current air quality standard (2008). The 1-hour exceedances for Simcoe and Essex are not available (MOE, 2007; Map base supplied Brock University Map Library).

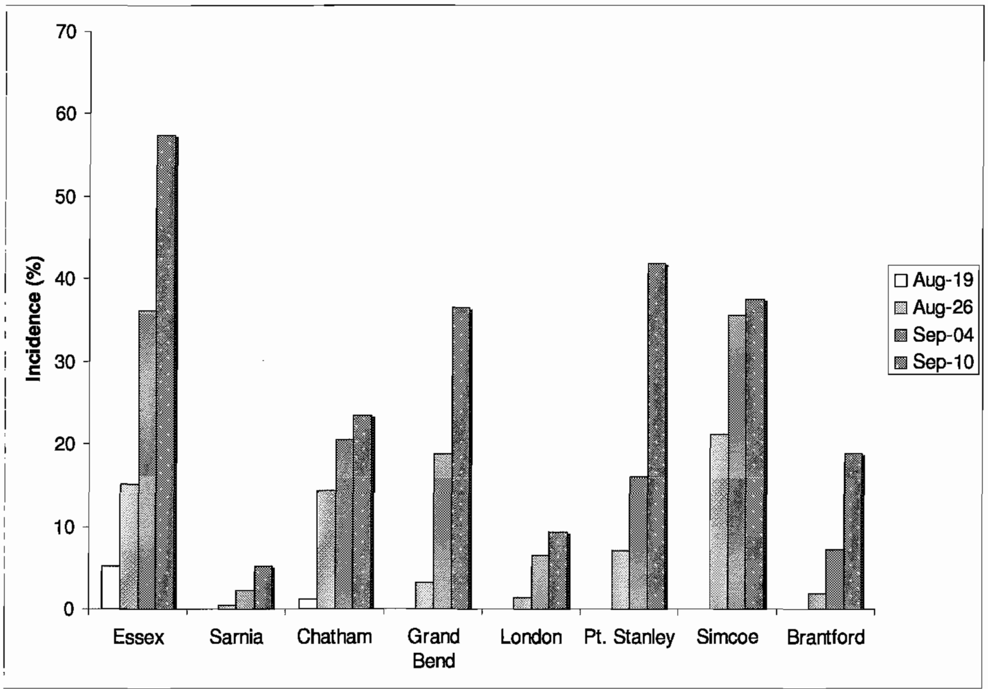


Figure 5.2: Incidence of milkweed leaves with visible ozone-induced damage defined by black stippling at seven survey sites in southern Ontario. By August 26 all sites showed some level of ozone-induced damage.

Table 5.2: Incidence of plants injured per site (%) at the last survey of the growing season (September 11, 2007) shown with cumulative O_3 exposure (ppb) and total daily precipitation (mm) taken from June 01-September 11). Simcoe had the highest O_3 exposure. Essex had the highest number of damaged plants and the most precipitation.

Site	**Incidence (plants/site)**	**Cumulative O_3 (ppb)**	**Total Daily Precipitation (mm)**
Essex	**89.33**	86741	**305.7**
Sarnia	36.00	82670	239.5
Chatham	36.00	94403	212.7
Grand Bend	53.33	83471	186.8
London	46.67	82277	186.4
Port Stanley	68.00	95251	164.2
Simcoe	76.00	96226	239.2
Brantford	46.67	83880	129.6

At the end of the season it was found that Chatham (1.84), Port Stanley (1.28) and Grand Bend (1.15) had the highest total O_3 values (units are Horsfall-Barratt Scale), followed by Essex (0.88), Simcoe (0.74), Brantford (0.54), Sarnia (0.25), and London (0.22).

5.3 Soil Moisture and Texture

The second driest spring on record occurred in 2007 in southern Ontario, and all study sites had lower than average total daily precipitation from June-September according to historical data, (Figure 5.3) (Environment Canada Climate Weather Office). Soil moisture was calculated per site from dry and wet weights of soil samples, (Table 5.3). The highest average seasonal (June-September) soil moisture were at Essex (32%), London (23%), Simcoe (22%) and Chatham (22%). This was followed by Port Stanley (20%), Brantford with (18%), Grand Bend (15%) and Sarnia (15%), (Figure 5.4). For the same period the highest total daily precipitation values were at Essex (311 mm), Sarnia (263 mm) and Simcoe (251 mm), followed by Chatham (221.7 mm), Grand Bend (213 mm), London (203 mm), Port Stanley (195 mm), and Brantford (165 mm).

Soil moisture and textures are found in Table 5.3. For the surveys conducted on June 9-11, August 19-21, August 26-28, September 04-05, and September 10-11 Essex had the highest soil moisture values. Grand Bend had the lowest soil moisture on three of the survey dates (August 06-08, August 26-68, and September 10-11).

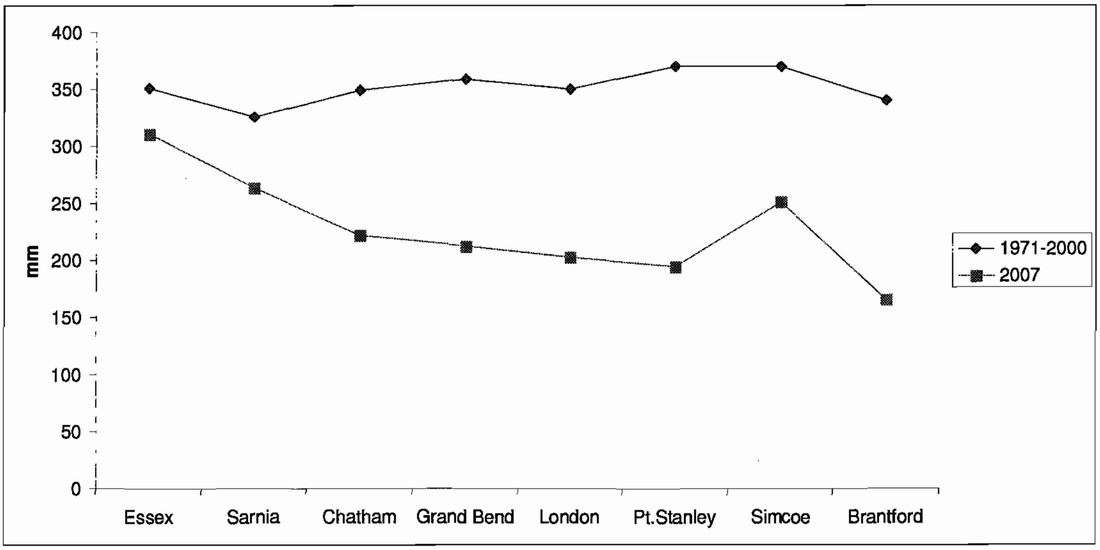


Figure 5.3: Total daily precipitation normals June-September (1971-2000) and the total daily precipitation from the survey months in 2007 (June-September). Retrieved on January 08, 2008 from http://climate.weatheroffice.ec.gc.ca/climateData/canada_e.html.

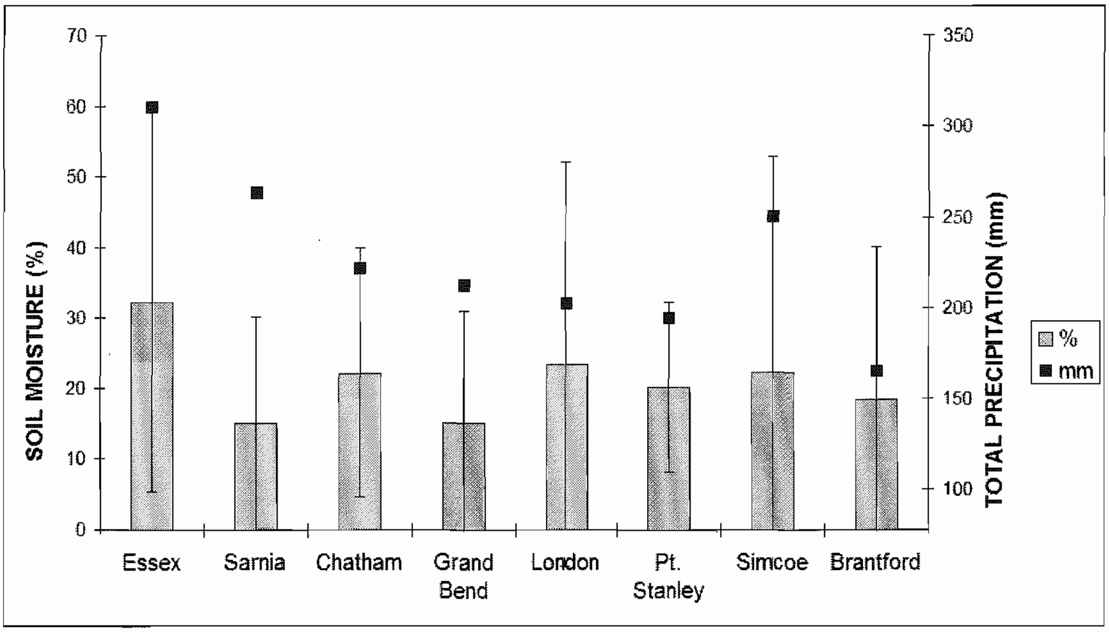


Figure 5.4: Average soil moisture values derived from soil samples taken on the seven survey dates are compared to total daily precipitation values retrieved from Environment Canada Climate Weather Office for the entire survey period (June 01-September 11). Bars showing standard deviation indicate the large variance in values. The average soil moisture calculations are not consistent with precipitation values.

Table 5.3: Measurements resulting from 25 g soil samples taken in 2007 at approximately 25 cm depth at each survey site (N=1). Soil textures are found in parentheses and highest soil moisture values are bold. Measurements: W=wet weight; D=dry weight in 100°C oven for 24 hours; % W= soil moisture.

Survey	June 9-11			July 17-20			August 06-08			August 19-21			August 26-28			September 04-05			September 10-11		
Measurement	W	D	% W	W	D	% W	W	D	% W	W	D	% W	W	D	% W	W	D	% W	W	D	% W
Site																					
Brantford (Clay)	28.97	17.47	66	13.06	12.64	3	29.85	27.55	8	28.74	27.15	6	29.83	24.99	19	24.41	22.64	7.8	33.89	28.80	18
London (Sand)	23.70	12.66	87	24.61	22.83	8	20.93	19.91	5	23.09	19.35	19	26.74	22.24	20	26.16	23.99	9.0	30.54	26.38	16
Grand Bend (Sand)	38.26	26.26	46	37.75	29.53	**28**	28.10	27.11	4	27.98	25.87	8	24.72	22.23	11	18.30	17.30	5.7	37.49	36.37	3
Sarnia (Silt)	34.40	23.71	45	37.02	35.98	3	24.13	22.87	6	17.46	15.13	15	31.72	29.02	10	26.15	25.28	3.4	26.51	25.46	4
Essex (Clay)	27.22	14.34	**90**	27.32	24.63	11	36.20	28.66	26	23.18	18.66	**24**	22.55	18.37	**22**	33.17	29.06	**14.2**	32.08	23.26	**38**
Chatham (Silt)	28.67	17.76	62	34.18	29.81	15	41.11	35.43	16	27.65	22.86	21	24.93	21.86	14	26.01	23.67	9.9	30.09	25.37	19
Pt. Stanley (Silt)	37.34	26.60	40	25.72	21.03	22	36.94	28.99	**27**	19.44	16.09	20	23.23	19.85	17	22.10	21.54	2.6	34.14	30.93	10
Simcoe (Clay)	24.22	12.64	92	19.93	18.41	8	25.96	23.50	11	23.95	21.62	10	31.92	28.14	13	27.40	25.24	8.6	28.35	24.99	13

5.4 Statistical Analysis

5.4.1 ANOVA

The results of the statistical analysis are found in Tables 5.4, 5.5 where a critical F-value greater than 4.2 and p-value of below 0.05 is statistically significant. Analysis of variance was conducted first to check for significance with each independent variable (SUM65 and cumulative O_3). The effect of SUM65 on the incidence of leaves per plant damaged by O_3 was significant with a value of $F(1,28)=16.49$, $p=0.0004$. The effect of total daily precipitation alone on the incidence of leaves per plant damaged by O_3 was significant with a value of $F(1,28)=7.41$,$p=0.0111$). Time and SUM65 were both highly significant to the incidence of leaves per plant damaged by O_3 ($F(1,28)=26.98$, $p=0.0001$). The effect of SUM65 alone on the incidence of plants damaged by O_3 was highly significant ($F(1,28)=17.37$, $p=0.0003$). The effect of total daily precipitation alone on the incidence of plants damaged by O_3 was also significant ($F(1,28)=5.32$, $p=0.0287$). The independent variables Time and SUM65 on incidence of plants damaged by O_3 was highly significant as well ($F(1,28)=13.72$, $p=0.0009$).

Table 5.4: Results of univariate analysis of variance test using the O_3 exposure index SUM65. Time indicates the repetition of the surveys (time1 = survey1) Statistical significance is achieved with an F-value greater than 4.2 and p-value of below 0.05 (bold).

Dependent Variable	Independent Variables	Results:
Incidence of Leaves Damaged	SUM65	**$F(1,28)=16.49$, $p=0.0004$**
	Total daily precipitation	**$F(1,28)=7.41$, $p=0.0111$**
	SUM65 * time	**$F(1,28)=26.98$, $p=0.0001$**
Incidence of Plants Damaged	SUM65	**$F(1,28)=17.37$, $p=0.0003$**
	Total daily precipitation	**$F(1,28)=5.32$, $p=0.0287$**
	SUM65 * time	**$F(1,28)=13.72$, $p=0.0009$**
Total O_3 Damage	SUM65	**$F(1,28)=4.28$, $p=0.0479$**
	Total daily precipitation	$F(1,28)=1.33$, $p=0.2593$
	SUM65 * time	**$F(1,28)=20.41$, $p=0.0001$**

Lastly, total O_3 damage was found to be significantly affected by SUM65 alone ($F(1,28)=4.28$, $p=0.0479$). Total daily precipitation did not significantly affect total O_3 damage ($F(1,28)=1.33$, $p=0.2593$). Time and SUM65 together were found to significantly affect the total O_3 damage as well ($F(1,28)=20.41$, $p=0.0001$).

The results of the ANOVA analysis for all three dependent variables can be found in Table 5.5 where a critical F-value greater than 4.2 and p-value of below 0.05 is statistically significant. The incidence of leaves damaged per plant was not significantly affected by cumulative O_3 ($F(1,28)=2.69$, $p=0.1122$). The incidence of leaves damaged per plant was significantly affected by total daily precipitation ($F(1,28)=7.38$, $p=0.0112$). Finally, the incidence of leaves damaged per plant was significantly affected by time and cumulative O_3 ($F(1,28)=29.34$, $p=0.0001$). The incidence of plants damaged per site was found to be significantly affected by cumulative O_3 alone ($F(1,28)=7.5$, $p=0.0106$). The effect of total daily precipitation on plants damaged per site was also significant ($F(1,28)=8.4$, $p=0.0072$) and the effect of time and cumulative O_3 on plants damaged per site produced a highly significant value of $F(1,28)=18.36$, $p=0.0002$.

Table 5.5: Results of univariate analysis of variance test using the exposure index cumulative O_3. Statistical significance is achieved with an F-value greater than 4.2 and p-value of below 0.05 (bold).

Dependent Variable	**Independent Variables**	**Results:**
Incidence of Leaves Damaged	Cumulative O_3	$F(1,28)=2.69$, $p=0.1122$
	Total daily precipitation	**$F(1,28)=7.38$, $p=0.0112$**
	Cumulative O_3 * time	**$F(1,28)=29.34$, $p=0.0001$**
Incidence of Plants Damaged	Cumulative O_3	**$F(1,28)=7.5$, $p=0.0106$**
	Total daily precipitation	$F(1,28)=8.4$, $p=0.0072$
	Cumulative O_3 * time	**$F(1,28)=18.36$, $p=0.0002$**
Total O_3 Damage	Cumulative O_3	$F(1,28)=0.36$, $p=0.5543$
	Total daily precipitation	$F(1,28)=0.16$, $p=0.6930$
	Cumulative O_3 * time	**$F(1,28)=19.51$, $p=0.0001$**

Lastly, total O_3 damage was not significantly affected by cumulative O_3 alone nor total daily precipitation alone with $F(1,28)=0.36$, p=0.5543and $F(1,28)=0.16$, p=0.6930, respectively. Time and cumulative O_3 together did produce highly significant results ($F(1,28)=19.51$, p=0.0001).

The results of the statistical analysis for mean daily air temperature effects can be found in Table 5.6 where a critical F-value greater than 5.59 and p-value of below 0.05 is statistically significant. Using SUM65 as the independent variable it was found that mean daily air temperature alone did not significantly affect the incidence of leaves damaged per plant ($F(1,7)=0.28$, p=0.6022), plants damaged per site ($F(1,7)=0.44$, p=0.5119) nor total O_3 ($F(1,7)=0.54$, p=0.4689). Again, the effect of mean daily air temperature alone with cumulative O_3 as the dependent variable was insignificant for the incidence of leaves damaged per plant ($F(0.34)$, p=0.5663), plants damaged per site ($F(1,7)= 76.24$, p=0.4859) and total O_3 ($F(1,7)=0.11$, p=0.4477). However, after adding mean daily air temperature to the model with total daily precipitation the independent variables were found to still significantly affect all three dependent variables for SUM65: leaves damaged per plant ($F(1,7)=9.81$, p=0.0039), plants damaged per site ($F(1,7)=12.04$, p=0.0016) and total O_3 ($F(1,7)=9.51$, p=0.0045), and for cumulative O_3 the values were $F(1,7)=4.18$, p=0.0500, $F(1,7)=11.88$, p=0.0018, and $F(1,7)=7.14$, p=0.0123 respectively. Because mean daily air temperature was insignificant itself and did not change the previous findings of significance for the dependent variables it was not used further in the analysis.

5.4.2 MANOVA

It was found that there was no site effect for incidence of leaves damaged per plant, plants damaged per site nor total O_3 with $F(7,32)=1.22$, p=0.3222, $F(7,32)=0.95$, p=0.4826and $F(7,32)=1.85$, p=0.1124, respectively (F-critical=2.3). All three dependent variables were then combined to look for site effect while reducing Type I error with a critical F-value of 1.7 the resulting $F(21,96)$,=1.62, p=0.0595 was again insignificant.

Table 5.6: Results of univariate analysis of variance test when mean daily air temperature was factored in. Statistical significance is achieved with an F-value greater than 5.59 and p-value of below 0.05 (bold).

Dependent Variable	Independent Variables	Results:
Incidence of Leaves Damaged	SUM65*Mean daily air temperature	$F(1,7)=0.28$, $p=0.6022$
	Cumulative O_3*Mean daily air temperature	$F(1,7)=0.34$, $p=0.5663$
	SUM65*Mean daily air temperature*Total daily precipitation	**$F(1,7)=9.81$, $p=0.0039$**
	Cumulative O_3* Mean daily air temperature*Total daily precipitation	$F(1,7)=4.18$, $p=0.0500$
Incidence of Plants Damaged	SUM65*Mean daily air temperature	$F(1,7)=0.44$, $p=0.5119$
	Cumulative O_3*Mean daily air temperature	$F(1,7)=0.50$, $p=0.4849$
	SUM65*Mean daily air temperature*Total daily precipitation	**$F(1,7)=12.04$, $p=0.0016$**
	Cumulative O_3 *Mean daily air temperature*Total daily precipitation	**$F(1,7)=11.88$, $p=0.0018$**
Total O_3 Damage	SUM65*Mean daily air temperature	$F(1,7)=0.54$, $p=0.4689$
	Cumulative O_3*Mean daily air temperature	$F(1,7)=0.11$, $p=0.4477$
	SUM65*Mean daily air temperature*Total daily precipitation	**$F(1,7)=9.51$, $p=0.0045$**
	Cumulative O_3 *Mean daily air temperature*Total daily precipitation	**$F(1,7)=7.14$, $p=0.0123$**

The next multivariate tests were to determine if there was an effect of site when adding the variables SUM65 and total daily precipitation. This test revealed that the effect of site along with SUM65 and total daily precipitation are significant $F(21,78)=3.72$, $p=0.0001$. Finally cumulative O_3 and total daily precipitation were added and tested for site effect which resulted in a value of $F(21,78)=1.98$, $p=0.0162$, again significant (F-critical 1.7).

5.5 Hypotheses

The first hypothesis is stated as H_0: *there is no identifiable relationship between O_3 exposure and milkweed O_3 damage.* Based on the statistical findings the H_0 is rejected in all cases stating that there is a significant relationship between SUM65 and all three indices of milkweed O_3 damage.

When using cumulative O_3 as the independent variable the H_0 is rejected for the incidence of plants damaged per site, stating that there is a significant relationship between cumulative O_3 and incidence of milkweed O_3 damage to plants per site. For the incidence of leaves damaged per plant and total O_3 the H_0 is not rejected stating that there is no identifiable relationship between cumulative O_3 exposure and incidence of leaves damaged nor total O_3 damage.

The second hypothesis is stated as H_0: *There is no effect of total daily precipitation on milkweed O_3 damage.* For the independent variable SUM65 the H_0 is rejected for leaves damaged and plants damaged, stating that there is a significant effect of total daily precipitation on the incidence of leaves damaged per plant, as there is a significant effect of total daily precipitation on the incidence of plants damaged per site. For total O_3 damage the H_0 was not rejected stating that there is no effect of total daily precipitation on the total O_3 damage to milkweed plants.

For the independent variable cumulative O_3 the H_0 is rejected for leaves damaged and plants damaged, stating that there is a significant effect of total daily precipitation on the incidence of leaves damaged per plant, as there is a significant effect of total daily precipitation on the incidence of plants damaged per site when cumulative O_3 is the independent variable. For total O_3 damage the H_0 was not rejected stating that there is no effect of total daily precipitation on the total O_3 damage to milkweed plants.

The third hypothesis is stated as H_0: *There is no effect of mean daily air temperature on milkweed O_3 damage.* For the independent variables SUM65 and cumulative O_3 the H_0 was not rejected stating that there was no effect of mean daily air temperature on all three types of milkweed O_3 damage.

The fourth hypothesis is stated as H_0: *There is no effect of survey location (site) on milkweed O_3 damage.* When testing for site effect alone the H_0 was not rejected stating that there is no effect of site alone on all three types of milkweed O_3 damage. When the analysis was adjusted for SUM65 and total daily precipitation, and cumulative O_3 and total daily precipitation the H_0 was rejected stating that there was a significant effect of site on all three types of milkweed O_3 damage when the independent variables and precipitation are factored into the analysis.

Chapter 6: Discussion

6.1 Ozone Exposure and Damage

6.1.1 Exposure Indices

The ozone monitoring community has not identified one single index to report all O_3 exposures because each one has different relevance. In this study eight O_3 indices were calculated to determine which was the best at indicating O_3 exposures that are biologically relevant in southern Ontario. It was found that the 12-hr and 24-hr averages were less accurate at summarizing the damaging effects of O_3. This is especially true for the 24-hr average because this included the evening hours when O_3 is low and damage is minimal because the stomata are closed. The 12-hr average only accounts for the daylight hours allowing for a more biologically useful value because the stomata are usually open for daytime respiration. Port Stanley had the highest 12-hr average, which is consistent with the 1-hr exceedances of 232 hours for 65 ppb and 66 hours for 80 ppb.

The N100 is not useful for statistical analysis as the values are often zero. Interestingly Grand Bend did not have high O_3 values overall, it did have acute highly elevated O_3 events. Grand Bend experiences heavy visitor traffic during the summer months, which adds primary pollutants to the air increasing the potential O_3 development, the combination of these two factors could be the cause of these O_3 events. One of the highest O_3 days for Grand Bend occurred on August 02, the start of the civic holiday weekend in Canada. The N100 gives a measure of the acute O_3 exposures that can damage sensitive plants. This index should not be used alone as it does not count the exposures between 60-100 ppb, which are known to cause O_3 damage to plants.

The SUM80 calculates the amount of O_3 recorded each hour above 80 ppb. This index is useful for identifying the sites that have elevated O_3 and are in non-attainment of the current O_3 standard of 80 ppb. The SUM80 only shows elevated values above 80. It is useful, but the biologically relevant levels of O_3 exposure (60-80 ppb) are not included in the summation. Both the N100 and SUM80 would need to be used in conjunction with another O_3 reporting index (e.g. W126 or cumulative O_3) in order to accurately characterize the O_3 exposure. This conclusion applies equally to all sites because plant damage begins before 80 ppb.

The most biologically relevant O_3 exposure indices were found to be cumulative O_3 and SUM65. Simcoe has the highest cumulative O_3, but Port Stanley had more hours above 65 and 80 ppb. However, even though Simcoe had more total O_3 it was low level O_3 (<65 ppb), which does not always produce plant damage. The SUM65 was used because 65 ppb is the level above which negative human and plant health effects begin (MOE, 2006). The SUM65 will also identify the sites where ambient O_3 levels are chronic at moderate levels. Port Stanley had many 65 ppb exceedances, but very few highly elevated exceedances (>100 ppb). SUM65 data show that while all sites may not have highly elevated O_3, they do all have slightly elevated O_3, which is responsible for most of the damage.

The backwards trajectories that were created for the highest O_3 days during the summer of 2007 showed what has long been known and is well documented. Southern Ontario receives transboundary air pollution originating in the U.S., which combines with local air pollution resulting in the highest O_3 levels in all of Canada, (Toronto Public Health, 2005; M.O.E., 2007). These results also show that wind direction is an important factor when considering O_3 exposure in southern Ontario.

6.1.2 Damage Indices

The actual ozone-induced damage to the milkweed plants was assessed and reported as four different parameters: the incidence of leaves damaged per plant, the incidence of plants damaged per site, the total O_3 damage and the Horsfall-Barratt scale of stipple cover. The purpose of reporting four different formats was to determine which method most accurately characterized ozone-induced damage when compared to known O_3 exposures. ANOVA tests revealed that three O_3 damage variables (incidence of plants damaged, incidence of leaves damaged, total O_3) were significantly related to SUM65 and the incidence of plants damaged per site was the most highly significant of all three. Each of the dependent variables measures O_3 damage at different echelons (leaf, plant, site) and because of this there was variation in the strength of each index.

The incidence of leaves damaged per plant calculates the damage per leaf. This shows the variation of leaf damage within each plant and results in a weak measurement when correlated to O_3 exposure making incidence of leaves damaged per plant a weak measurement.

The incidence of leaves damaged per plant gives an indication of the health of each individual plant at the site at the end of the season. Essex had the highest average number of leaves damaged followed by Port Stanley and Simcoe. This occurred even though Port Stanley and Simcoe were the sites with the highest O_3 exposure and Essex had the fourth highest O_3 exposure of eight sites. Essex had 311 mm of rain during the O_3 season, where Port Stanley had the least of all eight sites, 194.5 mm. Even though Port Stanley has 232 hours above 65 ppb and Essex had 150, Essex still has more leaf damage possibly because moisture is a significant variable in allowing plant gas exchange. The stomata may close due to low soil moisture, which may have been the case at Port Stanley.

The number of plants damaged per site gives a more precise characterization than the number of leaves damaged or the total O_3 because there is less variation when surveying at the plant level. When a plant shows any amount of ozone-induced damage the plant is counted as injured and this measure was most strongly correlated to actual O_3 exposure. When assessing the plants damaged per site each leaf is not surveyed and the variation that is inherent in each individual plant is absent in this index making it the stronger reporting parameter. At the end of the season Essex had the most plants damaged per site. When comparing simply O_3 exposure to O_3 damage this might be unexpected because Essex did not have the highest O_3 exposure. However, when accounting for the total daily precipitation, which allows for normal plant function including gaseous uptake, these results make sense. Simcoe had the second largest number of plants damaged, which was expected because Simcoe had one of the highest O_3 values. Port Stanley had the third largest number of plants injured and one of the highest O_3 values, but as previously mentioned had the lowest total daily precipitation, which may be why it was third of the high O_3 sites.

Total O_3 damage was determined by multiplying the average Horsfall-Barratt value by the incidence of leaves injured per plant to arrive at a value that is representative of the whole plant and not just the injured or non-injured leaves. The total O_3 damage was found to be a more accurate measure of the damage per plant than the Horsfall-Barratt scale alone, however, the total O_3 calculation is based on the Horsfall-Barratt scale. Again Chatham had the highest level of leaf damage (total O_3) based on few severely damaged leaves, when actually only 23% of the leaves per plant were damaged and only 36% of the plants at the site were damaged.

Total O_3 was found to be the weakest dependent variable when tested with SUM65 as the independent variable, total O_3 damage was insignificant $F(1,28)=0.36$, $p=0.5543$). This could be because the index is calculated from the Horsfall-Barratt scale, which assigns a percent cover value to each leaf set. The percent damage cover is then averaged for all leaf sets and multiplied by the probability of injury occurring per plant (incidence). This measurement can become less representative when the distribution of damage to each plant is inconsistent, or highly variable. This measure was used here because it is useful for describing O_3 damage and how it effects each individual plant, however, it is not the strongest at describing the O_3 damage to the whole site. When assessing O_3 at the individual plant level the individual leaf resistance, and microclimatic conditions can result in some variability. The Horsfall-Barratt scale was significantly related to O_3 exposure, however it was not the strongest measure of O_3 injury and it was not used to describe the actual duration or timing of the O_3 exposure.

6.1.3 Horsfall-Barratt Methodology

In this case study a Horsfall-Barratt value was determined for each leaf (up to 36 per plant) and then a value was assigned to each plant by taking an average of mid-point of each class for all of the assigned leaf values. This resulted in a weak statistical approach and values that did not accurately describe the damage to the whole site because the percentage of damage was averaged with the non-damaged leaves. The Horsfall-Barratt scale is useful in describing the percent of leaf surface covered with damage but when taken alone turned out to be the weakest reporting parameter in terms of characterizing the whole site. It was determined that injury per plant would be the best measure, not the percentage of injury on each leaf. Because there are natural variations within each plant this turned out to be the weakest parameter and subsequently not tested statistically, but it was chosen as part of the methodology in order to determine which damage parameter was most efficient. The results show that when characterizing a whole site the Horsfall-Barratt scale took longer than the other measures and was not as accurately related to actual O_3 exposures. Using this methodology wasted a lot of time, as it was the most difficult to complete and was the weakest statistically. The Horsfall-Barratt scale would be useful for studies that have repeated surveys on the same plant using exact doses of O_3 in order to closely monitor the damage or in a laboratory setting where extensive leaf analysis could be conducted (Duchelle & Skelly, 1981).

6.1.4 Meteorological Variables

For most of southern Ontario the temperature was 2-3°C above average and the total daily precipitation during the spring months was 40-85% of climatic norms (N.A.D.M, 2007). In the summer months some areas had only 10-40% of normal precipitation (N.A.D.M, 2007). Mean daily air temperature was also factored into the MANOVA analysis to test if there was an effect. Temperature by itself was found to have no significant effect on all three O_3 damage variables and adding mean daily air temperature did not change the significance of the previous tests. It was for this reason that the mean daily air temperature was tested using MANOVA, but not entered into the models after it was determined to have no effect. Although mean daily air temperature is expected to have an effect on atmospheric O_3 production, soil moisture, and plant moisture status it was not correlated to plant damage. When total daily precipitation and mean daily air temperature were assessed it was found that precipitation was the only one correlated to ozone-induced damage. This is because moisture must be present for the stomata to remain open so that gas exchange can occur in the presence of elevated O_3.

6.1.5 Survey Site Variation

Multivariate analysis showed that the geographic location of the milkweed (site) alone was not significantly related to the three types of ozone-induced damage, which showed that damage was instead related to O_3 exposure and total daily precipitation. SUM65 and cumulative O_3 were added independently into the model in two different tests with total daily precipitation and it was found that indeed the effect of these variables on ozone-induced damage was significant and correlated to site. This suggests that there was some spatial variability in ozone-induced plant damage within and between sites. This variability may reflect intra and inter-site variability in O_3 exposure, total daily precipitation and damage.

6.2 Soil Moisture and Texture

Soil moisture was compared to O_3 exposures to determine how these two factors contribute to producing ozone-induced damage to milkweed plants. The soil collections were made during surveys that were conducted monthly (May, June, July) and weekly (August, September). Because of this periodic sampling of the soil, water content calculations did not accurately characterize the actual soil moisture. For this reason the actual amount of total daily precipitation (mm) obtained from the Environment Canada Climate Weather Office was used in the statistical analysis. Total daily precipitation values showed that for the months June-September Essex had received by far (305.7 mm) the largest amount of rain of all the survey sites. Port Stanley, which is the site with the highest O_3 exposures received the lowest amount of rain with only 165.4 mm, which is 53% of the rain Essex received, all sites in 2007 experienced below average precipitation. Soil moisture is such an important factor in producing ozone-induced damage these levels effected how much damage was found on the plants.

The soil texture was determined by the Texture by Feel Method after Thien, S.J. (Kansas State University, 1979) to see if there was a relationship between texture and the soil moisture that characterized the growing conditions of the plants. The amount of water available in the soil can be in small part determined by its texture and this could therefore, affect the amount of water available for the plant to uptake. Three of the sites, Essex, Simcoe and Brantford had clay soil and these sites were expected to have some of the highest soil moistures because clay has a high water holding capacity. This was true for Essex and Simcoe, which had the highest soil moisture values but Brantford had one of the lowest soil moisture values, which was unexpected and could have been due to the drainage of the site, which was not taken into account for this study. All of the sites were on relatively flat ground with no slope, however, the drainage patterns of the sites were not assessed. Chatham, Pt. Stanley and Sarnia had silty soil. These three sites had high, medium and low soil moisture levels, which is not consistent for one soil type indicating that there was more involved in the water holding capacity of the soils than the texture. Grand Bend had a sandy soil, which allows water to travel through quickly and will have a lower water holding potential. As expected, Grand Bend had one of the lowest soil moisture values. It is clear that there is no identifiable relationship between apparent soil texture and soil moisture in this study because the soil moisture values obtained from the infrequent sampling may not have accurately represented the moisture status. The soil moisture measures may have been adequate if the sampling was done more frequently. Additionally, soil moisture is also affected by organic matter content, compaction, soil structure, plant cover type and actual evapotranspiration. In the future a more sophisticated means of characterizing the soil would result in more robust data.

6.3 Hypotheses

The hypothesis H_0: *there is no identifiable relationship between O_3 exposure and milkweed O_3 damage* was rejected in all cases except for the effect of cumulative O_3 on leaves damaged per plant and total O_3. Large variation occurs when sampling each leaf of the milkweed plants and this was the cause of these measures being weak. In the future when studying a park or field it is suggested to simply count the number of affected plants per site. This parameter is actually the most time effective as it does not involve determining a percent cover for each leaf in the area. It is important to note that ozone-induced damage can be correlated to O_3 levels but in this study area it was not used to describe the duration or timing of the O_3 exposure.

The hypothesis H_0: *There is no effect of total daily precipitation on milkweed O_3 damage* was rejected in all cases except for the effect of total daily precipitation on total O_3 damage. Total O_3 damage was the weakest variable of plant damage. For the incidence of plants and incidence of leaves damages total daily precipitation was shown to be significantly related to ozone-induced damage and is a necessary variable for this type of study.

Mean daily air temperature plays a role in atmospheric O_3 development and was expected to correlate with damage. It can only be ruled out that mean daily air temperature in so far as the specific variables that were chosen has low explanatory power. The H_0: *There is no effect of mean daily air temperature on milkweed O_3 damage* was not rejected in all cases. For the milkweed plants surveyed in this study the variables that were significantly correlated to the damage produced were O_3 exposure and total daily precipitation.

Finally, site effect alone was not associated with the production of ozone-induced damage, and did not change the significant values for the other O_3 damage tests. Ozone-induced damage was correlated to O_3 exposure and total daily precipitation at each site, and each site was significantly different from one another.

6.4 Conclusion

The objectives of this study were to systematically document milkweed damage, to explore the link between damage and O_3 exposure and to statistically characterize the relationship between mean daily air temperature, total daily precipitation and ozone-induced damage to milkweed in southern Ontario. Ozone-induced damage was found at eight sites in Ontario that have historically had high ambient O_3 levels. Statistical analysis of the SUM65 index and the dependent variables (incidence of leaves damaged, incidence of plants damaged and total O_3), showed the strongest association between the total number of injured plants per site, the SUM65 index, and total daily precipitation. Ozone-induced injury thresholds were not determined in this study as this type of measurement is most accurately determined in a controlled-exposure laboratory setting.

This study has shown that a statistical relationship exists between plant damage and precipitation data. This suggests that moisture plays an important role in expression of plant damage. If the moisture levels are known then a greater understanding of plant uptake is possible. Milkweed plants as bio-indicators can be useful for discovering areas that may need continuous monitoring using mechanical sensors, or to act as an early warning for other less sensitive plants and crops. The strength of bio-indicators is that they are a cost effective way to show the complexity of how ambient air pollution can effect biological systems beyond a simple ppb value that is produced from a sensor. In order to develop meaningful regulations it is imperative that the effects of O_3 on our environment are understood as completely as possible.

This study revealed that standard biomonitoring methodologies can be used to determine the extent of O_3 levels in the O_3 rich part of Ontario. These O_3 levels are having an impact on milkweed plants and very little is known about the significance of the impact on milkweed and the organisms that interact with milkweed. A protocol similar to this one could be utilized anywhere milkweed is found as a preliminary assessment of the O_3 exposure in that region, this could be the foundation to building a case requesting for more monitoring in areas that lack sensors.

6.5 Suggestions For Future Study

For future studies it is suggested from these findings that the soil moisture be more accurately characterized possibly using a data logger, which would be placed at the center of each site and a more thorough daily assessment of the soil moisture status and mean daily air temperature could be determined. This could more accurately address the hypotheses dealing with the O_3 damage and soil moisture. The reader is referred to Bergweiler *et al.* (2008) who monitored temperature, humidity, and photosynthetically active radiation. In that study net photosynthesis and stomatal conductance were directly measured, relieving the need for surrogate measurements of soil moisture. It is also possible that an evaporation pan could be used, which measures the amount of rainfall and subsequent evaporation at each site, which would give an indication of the moisture that is available to the plants. The use of a data logger and an evaporation pan requires that the site is open grown with no buildings nearby, flat and protected from vandals, landscaping maintenance, and animals. Another methodology could assess the actual plant moisture status or potential evapotranspiration, which may give the most accurate measures of plant moisture status (Bergweiler *et al.*, 2008). If the soil moisture could be more accurately determined using better technology or sampling methods it might not be necessary to know the soil textures in order to understand the site specific differences in water regime of the soil and therefore the plant.

Simply counting the number of plants showing damage would save time and allow for an increase in the number of plants surveyed, as this measurement was the strongest assessment and happens to be the easiest. By adopting this methodology, as seen in Davis & Orendovici (2006), the number of plants surveyed could be doubled. It would also be interesting to repeat a similar study with labeled plants at a protected site, visited repeatedly throughout the study period, which would give a better understanding of the individual plants response to O_3 exposure and possibly answer some of the questions about individual variation. Also, if a site is protected, it could be assessed year after year for analysis.

It was found that taking many measurements per plant (12-36) did not increase the value of the data, it would be more time-effective and create more robust data to simply survey more plants and record less information. In this case simply measuring the presence or absence of O_3-induced damage would be most useful for future study. The National Park Service of the U.S. has identified over 60 plant species that are known to be sensitive to O_3 exposure (NPS, 2003). Additionally, Canadian researchers are aware of over a dozen crop species that are sensitive to O_3 exposure (Pearson, 1992). Future studies could not only increase the number of milkweed surveyed by indicating presence or absence of O_3-induced damage, but could also utilize other plants and crops as indicators. This type of data would not only indicate O_3 presence, but also could implicate O_3 exposure in crop yield reduction.

The possible negative impact of O_3 exposure on the genetics and reproduction potential of milkweed was not investigated for this study. However, the presence of O_3 is thought to cause a reduction in seed pods, which could result in lower genetic variability and fewer milkweed plants. Few studies have thoroughly examined these issues in milkweed (Farnsworth, 2001; Bennett *et al.*, 2006). It would be useful to determine the exact outcome of O_3 exposure on milkweed populations and reproduction potential. This would provide additional information as to the long-term affects of O_3 exposure beyond the seasonal injury.

The possible ecosystem effects of milkweed reduction or total loss were not estimated. A greater understanding of milkweed importance to its surrounding plants and insects may reveal that milkweed reductions could result in ecosystem imbalance.

In particular monarch butterflies migrate across North America feeding on milkweed plants acquiring Cardenolides from their leaves, which are required for monarch growth and development (Malcolm & Zalucki, 1999). The monarch's survival depends on the health of the milkweed plant. A reduction in naturally occurring milkweed due to O_3 exposure could have negative effects on the monarch migration and possibly reduce the populations. These factors outline the importance of using plants as bio-indicators. The reader is referred to Zalucki *et al.*, 2002 for monarch-milkweed interactions.

Finally, the nutrient status of the soil was not assessed and it would be interesting to determine if each site has similar nutrients for plant uptake. Because soil nutrients may affect plant health it would be interesting to determine if this also affects O_3 injury. This could be done by testing the soil samples for nutrients, organic matter, and elemental content (see Bennett *et al.*, 2006; U.S.D.A. Soil Survey Reports).

In conclusion I found this study of ozone-induced milkweed damage to be a success and a good starting point for Ontario milkweed-ozone studies. The data collected here is representative of only the elevated O_3 regions of Ontario and of only one growth season. For this study many parameters and methodologies were assessed to determine which ones were most accurate for ozone-milkweed surveys. The purpose of testing the different methodologies used by the plant biomonitoring community was to assess if there is a "best" method. While sections of this work will need further investigation I believe that some of the conclusions will help future researchers optimize their time and effort. In the end it was affirmed that milkweed plants are indeed reliable indicators of O_3 exposure but that the process of using *in situ* plants as indicators of atmospheric pollution presents a level of complexity beyond simply associating damage to exposure. This study is valuable because the previously used methodologies were tested in southern Ontario and lessons were learned about how to continue assessing ozone-induced damage using milkweed in an accurate and efficient manner.

References Cited:

Bargagli, R. Trace Elements in Terrestrial Plants: An Ecophysiological Approach to Biomonitoring and Biorecovery. Georgetown, TX, USA.: Springer-Varlag Berlin Heidelberg and R.G. Landes Company. 1998

Bell, M.L., McDermott, A., Zeger, S.L., Samet, J.M., Dominici, F. 004. Ozone and short-term mortality in 95 US urban communities, 1987-2000. Journal of American Medical Association **292**(19): 2372-2378

Bennett, J.P., Jepsen, E.A., Roth J.A. 2006. Field responses of *Prunus serotina* and *Asclepias syriaca* to ozone around southern Lake Michigan. Environmental Pollution **142**: 354-366

Bergweiler, C., Manning, W.J., Chevone, B.I. 2008. Seasonal and diurnal gas exchange differences in ozone-sensitive common milkweed (*Asclepias syriaca* L.) in relation to ozone uptake. Environmental Pollution **152**: 403-415.

Brock University Map Library: *Modified from:* St. Catharines [computer file]. (no date). St. Catharines, Ontario: Brock University Map Library. Available: Brock University Map Library Controlled Access http://www.brocku.ca/maplibrary/images/stcathv8.jpg. (Accessed October 14, 2008).

C.E.P.A. 1999. Canadian Environmental Protection Act, 1999. Retrieved 2007 from: http://www.ec.gc.ca/CEPARegistry/the_act/

Chappelka, A., Renfro, J., Somers, G., Nash, B. 1997. Evaluation of Ozone Injury on Foliage of Black Cherry (*Prunus serotina*) and Tall Milkweed (*Asclepias exaltata)* in Great Smoky Mountains National Park. Environmental Pollution **95**: 13-18

Climate Change 2001: The Scientific Basis: Intergovernmental Panel on Climate Change. Retrieved on March 07, 2007 from: http://www.grida.no/climate/ipcc_tar/wg1/index.htm

Davis, D.D., Orendovici, T. 2006. Incidence of ozone symptoms on vegetation within a National Wildlife Refuge in New Jersey, USA. Environmental Pollution **143**: 555-564

Duchelle, S.F., Skelly, J.M. 1981. Response of Common Milkweed to Oxidant Air Pollution in the Shenandoah National Park in Virginia. Plant Disease **65**: 661-663

Encyclopedia Britannica, 2007. Leaf Retrieved on May 03, 2007 from Encyclopedia Britannica Online http://www.britannica.com/ebc/art-66079/Structures-of-a-leaf

Environment Canada: Air Quality Service. Retrieved on February 02, 2008 from http://www.msc-smc.ec.gc.ca/aq_smog/on/on_e.cfm

Environment Canada Climate Weather Office. Retrieved on January 08, 2008 from: http://climate.weatheroffice.ec.gc.ca/climateData/canada_e.html

Environment Canada, 2007: Retrieved March 2008 from: http://www.ec.gc.ca/default.asp?lang=En&n=714D9AAE-1&news=C33CCDD9-3186-4A43-875A-A1D88135164F#_ftnref1

Faoro, F., Iriti, M. 2005. Cell Death Behind Invisible Sysmptoms: Early Diagnosis of Ozone Injury. Biologia Plantarum **49** (4): 585-592

Fiore, A., Jacob, D.J., Liu, H., Yantosca, R.M., Fairlie,T.D. & Li,Q. 2003. Variability in Surface Ozone Background Over The United States: Implications for Air Qualilty Policy. Journal of Geophysical Research **108** (D24): 4787

Griffiths, Heather. 2003. Revision of Factsheet *Air Pollution on Agricultural Crops*, Order No. 85-002. Prepared for Integrated Pest Management

Gunthardt-Goerg, M.S., McQuattie, C.J., Scheidegger, C., Rhiner, C., Matyssek, R. 2000.Ozone-induced cytochemical and ultrastructural changes in leaf mesophyll cell walls. Canadian Journal of Forest Research **27**: 453-463

Gunthardt-Goerg, M.S., McQuattie, C.J., Maurer, S., Frey, B. 2000. Visible and microscopic injury in leaves of five deciduous tree species related to current critical ozone levels. Environmental Pollution **164**: 243-266

Horsfall, J.G., Barratt, R.W. 1945. An improved grading system for measuring plant disease. Phytopathology **35**: 655

Hunsaker, C.T., Carpenter, D., Messer, J. 1990. Ecological indicators for regional monitoring. Bulletin of the Ecological Society of America **71**: 165-172

Iqbal, M., Abdin, M.Z., Mahmooduzzafar, Yunus, M., Agrawal, M. 1996. Resistance Mechanisms in Plants Against Air Pollution. In Yunus, M. and Iqbal, M. (Eds.) Plant Response to Air Pollution. John Wiley & Sons Ltd.

Kelly, N.A., Ferman, M.A., Wolff, G.T. 1986. The chemical and meteorological conditions associated with high and low ozone concentrations in southeastern Michigan and nearby areas of Ontario. Journal of Air Pollution Control Associated **36**: 150-158. As cited in US EPA, 1996. p 3-49

Kelly, N.A., Gunst, R.F. 1990. Response of ozone to changes in hydrocarbon and nitrogen oxide smog chambers filled with Los Angeles air. Atmospheric Environment **24**(12): 2991-3005. As cited in US EPA, 1996 p.3.49

Kohut, R. 2005. Handbook for Assessment of Foliar Injury on Vegetation in the National Parks. National Parks Service: USDOI

Kouterick, K.B., Skelly, J.M., Frederickesen, T.S., Steiner, K.C., Kolb, T.E., Ferdinand, J.A. 2000. Foliar injury, leaf gas exchange and biomass responses of black cherry (*Prunus serotina*) half-sibling families to ozone exposure. Environmental Pollution **107**: 117-126

Kricke, R., Loppi.S. 2002. Nimis, P.L., Scheidegger, C., Wolseley, P.A. (Eds.) In Monitoring with Lichens-Monitoring with Lichens. NATO Science Series: IV. Earth and Environmental Sciences-Vol.7. Netherlands: Kluwer Academic Publishers. 2002

Lee, J.C., Steiner, K.C., Zhang, J.W. Skelly, J.M. 2002. Heritability of ozone sensitivity in open-pollinated families of black cherry (*Prunus serotina* Ehrh.) Forensic Science **48**: 111-117

Malcolm, S.B., and M.P. Zalucki. 1999. Plant Latex and First-Instar Monarch Larval Growth and Survival on Three North American Milkweed Species. Journal of Chemical Ecology **25**:1827-1842

Manning, W.J., 1996. The Use of Plants as Bioindicators of Ozone. Retrieved 2007 from: http://www.fs.fed.us/psw/publications/documents/psw_gtr166/psw_gtr166_001_manning.pdf

Manning, W.J., Krupa, S.V., Bergweiler, C.J., Nelson, K.I. 1996. Ambient ozone in three Class I wilderness areas in the northeastern USA: Measurements with Ogawa passive samplers. Environmental Pollution **91**: 399-403

Manning, W.J. 2003. Detecting plant effects is necessary to give biological significance to ambient ozone monitoring data and predictive ozone standards. Environmental Pollution **126**: 375-379

Markert, B.A., 2003. Markert, B.A., Breure, A.M., Zechmeister, H.G. (Eds.) In Trace Metals and other Contaminants in the Environment Vol 6: Bioindicators and Biomonitors. Elsevier Science Ltd. Oxford

McCurdy, T.R. 1994. Concentrations of ozone in the lower troposphere (ambient air). In McKee, D.J. (Ed), Tropospheric Ozone: Human health and Agricultural Impacts (p.19-37) Boca Raton: CRC Press

McKee, D.J. 1994. Introduction. In McKee, D.J. (Ed.). Tropospheric Ozone: Human health and Agricultural Impacts. (p.3-8) Boca Raton: CRC Press

McKendry, I. 2004. Background Concentrations of $PM_{2.5}$ and Ozone in British Columbia, Canada. Retrieved 2007 from: www.env.gov.bc.ca/air/airquality/.../**background**_pm25_**ozone**.pdf

MOE Ministry of the Environment: Air Quality Index Data. Retrieved 2007 from: http://www.ene.gov.on.ca/en/air/aqo/index.php

MOE 1989. Impact of Ozone Exposure on Vegetation in Ontario. Prepared by R.G. Pearson for the Phytoxicology Section, Air Resources Branch, Ontario Ministry of the Environment.

MOE 2006. Ministry of the Environment: Air Quality in Ontario Report 2005.

MOE 2007. Ministry of the Environment: Air Quality in Ontario Report 2006.

Morgan, P.B., Ainsworth, E.A., Long, S.P. 2003. How does elevated ozone impact soybean? A meta-analysis of photosynthesis, growth and yield. Plant, Cell, Environment **26**: 1317-1328

Mustafa, M.G. 1990. Biochemical Basis of Ozone Toxicity. Free Radical Biology and Medicine **9**: 245-265

NASA: Earth Observatory: Chemistry in the sunlight. Retrieved 2008 from: http://earthobservatory.nasa.gov/Library/ChemistrySunlight/chemistry_sunlight3.html

NASA Website: Goddard Space Center. Atmospheric Ozone Data and Resources. Retrieved 2008 from: http://jcbmac.chem.brown.edu/myl/ct7/ozone/ozone_cycle.html

N.P.S., 2003. Ozone Sensitive Plant Species on National Park Service and U.S. Fish and Wildlife Service Lands: Results of a June 24-25, 2003 Workshop. Prepared by Ellen Porter, Air Resources Division, National Park Service. NPS/NRARD/NRR-2003/01. Retrieved September 2007 from www2.nature.nps.gov/air/Pubs/pdf/BaltFinalReport1.pdf

National Air Pollution Surveillance (NAPS) Network. Air Quality in Canada: 2001 Summary and 1990-2001 Trend Analysis.

Nimis, P.L., Scheidegger, C., Wolseley, P.A. (Eds.) In Monitoring with Lichens-Monitoring with Lichens. NATO Science Series: IV. Earth and Environmental Sciences-Vol.7. Netherlands: Kluwer Academic Publishers. 2002

Neufeld, H. S., Renfro, J. R., Hacker, W. D., Silsbee, D., 1992. Ozone in Great Smoky Mountains National Park: dynamics and effects on plants. In: Berglund, R. L. (Ed.) Tropospheric ozone and the environment II: effects, modeling and control [papers from an international specialty conference]; November; Atlanta, GA; Pittsburgh, PA: Air & Waste Management Association; pp. 594-617.

Neufeld, H.S., Chappelka, A.H., Somers, G.L., Burkey, K.O., Davison, A.W., Finkelstein, P.L. 2006. Visible foliar injury caused by ozone alters the relationship between SPAD meter readings and chlorophyll concentrations in cutleaf coneflower. Photosynthesis Research **87**: 281-286

Nolle, M., Ellul, R., Heinrich, G., Gusten, H. 2002. A long-term study of background ozone concentrations in the central Mediterranean-diurnal and seasonal variations on the island of Gozo. Atmospheric Environment **36:** 1391-1402.

Pearson, R.G. 1992. Impact of Ozone Exposure on Vegetation in Ontario. Prepared for the Phytoxicology Section, Air Resources Branch, Ontario Ministry of the Environment

Pell, E.J and Dann, M.S. 1991. Multiple Stress-Induced Foliar Senescence and Implications for Whole-Plant Longevity. In Mooney, H.A., Winner, W.E., Pell, E.J., Chu, E. (Eds.) Response of Plants to Multiple Stresses. (p.189-204) Toronto: Academic Press Inc.

Pell, E.J., Schlagnhaufer, C.D., Arteca, R.N. 1997. Ozone-induced oxidative stress: Mechanisms of action and reaction. Physiologia Plantarum **100**: 264-273

Pell, E.J., Sinn, J.P., Brendley, B.W., Samuelson, L., Vinten-Johansen, C., Tien, M., Slillman, J. 1999. Differential response of four tree species to ozone-induced acceleration of foliar senescence. Plant, Cell, Environment **22**: 779-790 http://www.airqualityontario.com/reports/historical.cfm

Plochl, M, Lyons, T., Ollerenshaw, J., Barnes, J. 2000. Simulating ozone detoxification in the leaf apoplast through the direct reaction with ascorbate. Planta **210**: 454-467

Rao, M.V., Koch, J.R., Davis, K.R. 2000. Ozone: a tool for probing programmed cell death in plants. Plant and Molecular Biology **44**: 345-358

Richards, B.L., Middleton, J.T., Hewitt, W.B. 1958. Air Pollution with relation to agronomic crops: V. Oxidant stipple of grape. Agronomy Journal **50**: 559-561

Saarela, L.-E., Harju, L., Rajander, J., Lill, J.-O., Heselius, S.-L., Lindroos, A., Mattsson, K. 2005. Elemental analyses of pine bark wood in an environmental study. Science of the Total Environment **343**: 231-241

Schaub, M., Skelly, J.M., Zhang, J.W., Ferdinand, J.A., Savage, J.E., Stevenson, R.E., Davis, D.D., Steiner, K.C. 2005. Physiological and foliar symptom response in the crowns of *Prunus serotina*, *Fraxinus Americana* and *Acer rubrum* canopy trees to ambient ozone under forest conditions. Environmental Pollution **133**: 553-567

Schraudner, M., Langebartels, C., Sanderman, H. 1997. changes in the biochemical status of plant cells induced by the environmental pollutant ozone. Physiologia Plantarum **100**: 274-280

SFSU, 2009: San Fransisco State University Biology Lab. Retrieved September 2009 from:
http://userwww.sfsu.edu/~biol240/labs/lab_01variation/pages/1significance.html

Skelly, J.M. 2000. Tropospheric ozone and its importance to forests and natural plant communities of the northeastern United States. Northeastern Naturalist **7**(3): 221-236

Somers, G.L., Chappelka, A.H., Rosseau, P., Renfro, J.R. 1998. Empirical evidence of growth decline related to visible ozone injury. Ecological Management **104**: 129-137

Souza, L., Neufeld, H.S., Chappelka, A.H., Burkey, K.O., Davison, A.W. 2006. Seasonal development of ozone-induced foliar injury on tall milkweed (*Asclepias exaltata*) in Great Smoky Mountains National Park. Environmental Pollution **141**: 175-183

Spellerberg, I. Monitoring Ecological Change Cambridge, UK ; New York : Cambridge University Press, 2005.

Tingey, D.T. and Taylor, G.E., Jr. 1982. Variation in Plant Response to Ozone: A Conceptual Model of Physiological Events. In Unsworth, M.H., Ormrod, D.P. (Eds.) Effects of Gaseous Air Pollution in Agriculture and Horticulture. (p.113-138). Toronto: Butterworth Scientific

Toronto Public Health, 2005. Curbing Transboundary Air Pollution: Protecting Health through Legal Action. A report from Dr. David McKewon, Medical Officer of Health. Toronto, Ontario

Torsethaugen G, Pell EJ, Assmann SM, 1999. Ozone inhibits guard cell K^+ channels implicated in stomatal opening. Proceeding of the National Academy of Sciences USA **96:** 13577–13582

US EPA, 1996 Air Quality Criteria for Ozone and Related Photochemical Oxidants. U.S. Environmental Protection Agency, Office of Research and Development, National Center for Environmental Assessment, Washington Office, Washington, DC, 1996. Retrieved 2007 from: http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=2831

US EPA, 2004. United States-Canada Air Quality Agreement Progress Report. 2004. EPA430-R-04-007. Retrieved 2007 from http://www.epa.gov/airmarkt/progsregs/usca/docs/airus04.pdf

U.S. E.P.A, 2006. Air Quality Criteria for Ozone and Related Photochemical Oxidants. vol I, II, III. U.S. Enviromental Protection Agency, Washington, D.C., EPA/600/R-05/004aF-cF, 2006. Retrieved 2008 from: http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=2831

Vollenweider, P., Ottiger, M., Gunthardt-Goerg, M.S. 2003. Validation of leaf ozone symptoms in natural vegetation using microscopical methods. Environmental Pollution **124**: 101-118

Wagner, J.G., Van Dyken, S.J., Hotchkiss, J.A., Harkema, J.R. 2002. Endotoxin Enhancement of Ozone-Induced Mucous Cell Metaplasia Is Neutrophil-Dependent in Rat Nasal Epithelium. Toxicological Sciences **60**: 338-247

Yap, D., Reid, N., De Brou, G., Bloxam, R. 2005. Transboundary Air Pollution in Ontario: Ministry of the Environment.

Yuska, D.E., Skelly, J.M., Ferdinand, J.A., Stevenson, R.E., Savage, J.E., Mulik, J.D., Hines, A. 2003. Use of bioindicators and passive sampling devices to evaluate ambient ozone concentrations in north central Pennsylvania. Environmental Pollution **125**: 71-80

Zalucki, M.P., Clarke, A.R., Malcom, S.B. 2002. Ecology and Behavior of First Instar Larval Lepidoptera. Annual Review of Entomology **47:** 361-393

Appendix I: Survey Sites

Brantford:

Type of location: Park (site was located along the walking trail approximately 100 meters after the entrance of Waterworks Park on Grand River Avenue)
Soil: Clay
Weather Station: Hamilton A

Chatham:

Type of location: Park (site was located along walking trail on 435 Grand Ave. W. road across from the government building)
Soil: Silt
Weather Station: Chatham WPCP

Essex:

Type of location: Field (site was located in the field across from the government building located at 360 Fairview Dr. W.)
Soil: Clay
Weather Station: Harrow CDA Auto

London:

Type of location: Park (site was located inside the Fanshaw Park, approximately 400 meters after the entrance on Fanshaw Park Road E)
Soil: Sand
Weather Station: London

Grand Bend:

Type of location: Trail (site was located along a hiking trail off of Lakeshore Road)
Soil: Sand
Weather Station: Exeter

Port Stanley:

Type of location: Field (site was located at the end of the Hawk's Cliff Road which intersects with Dexter Road, near the cliffs)
Soil: Silt

Sarnia:

Type of location: Park (site was located along walking trails off of Exmouth St. approximately 400 m from the intersection of Front St., near Centennial Park)
Soil: Silt
Weather Station: Sarnia Climate

Simcoe:

Type of location: Field (site was located in a field across from an apple farm on Concession 6/Woodhouse road approximately 500 meters from the intersection of Blue Line Rd and Concession 6)
Soil: Clay
Weather Station: Ridgetown

Appendix II: Images of Ozone-induced Damage and Other Types of Damage Not Related to Ozone

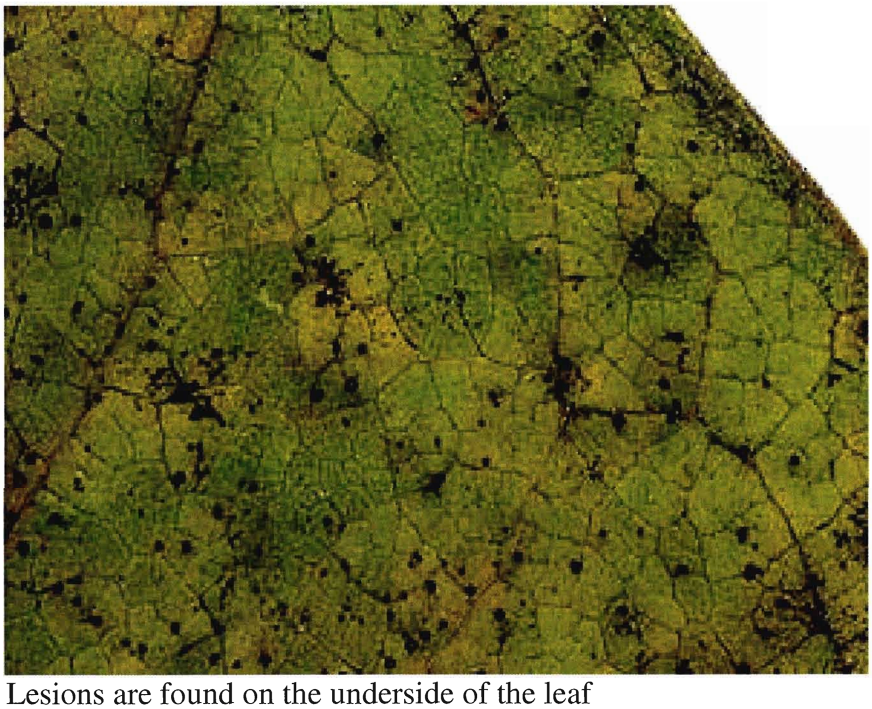
http://www.dnr.state.wi.us/org/caer/ce/eek/earth/field/milkweed/slideshowindex.htm

Non-damaged Milkweed Leaf

O_3-induced Stippling

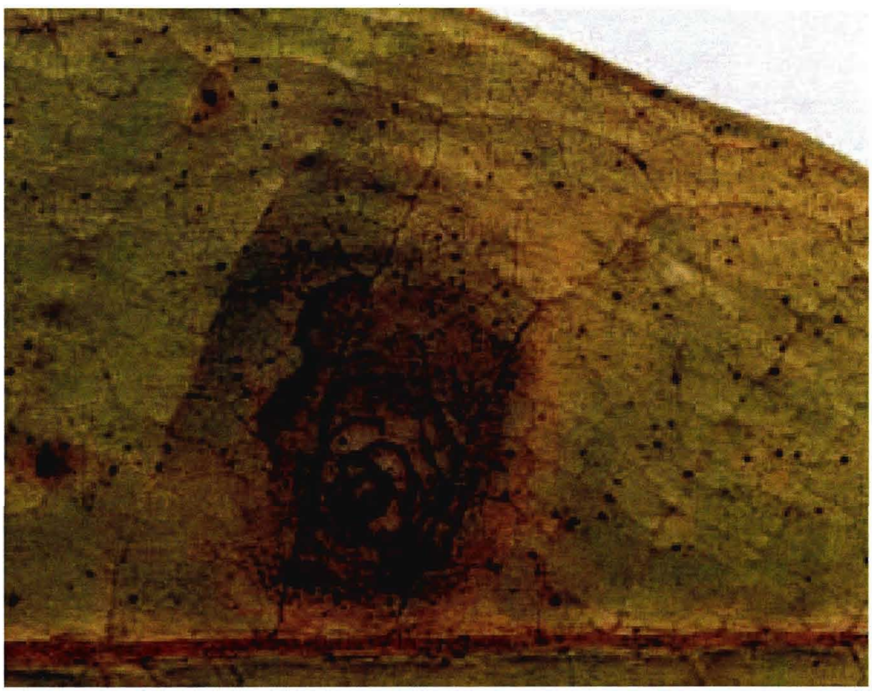
Damage to Milkweed Plants Not Related to Ozone Exposure

Lesions

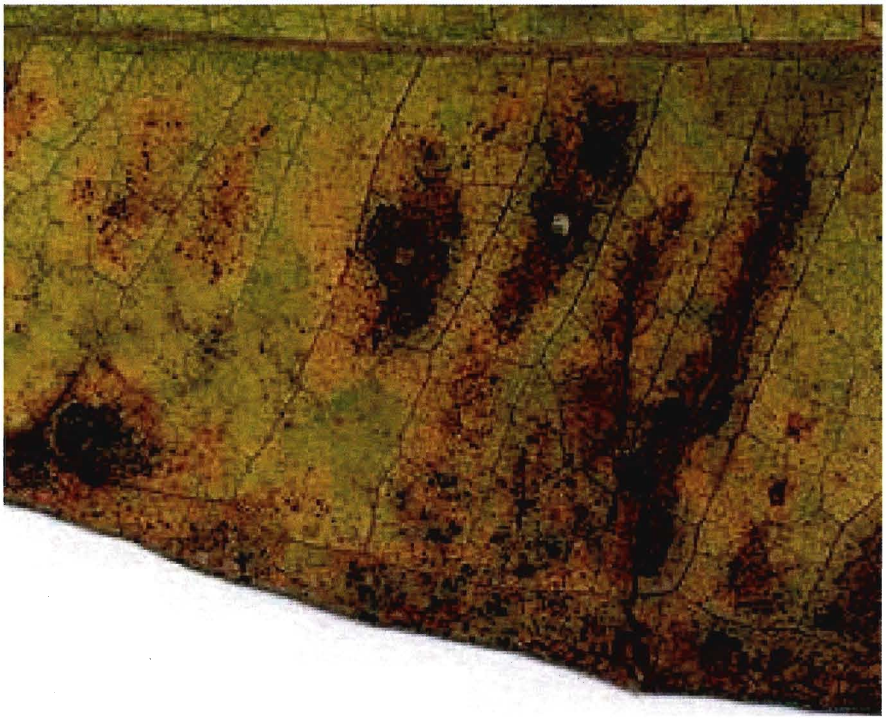


Lesions are found on the underside of the leaf

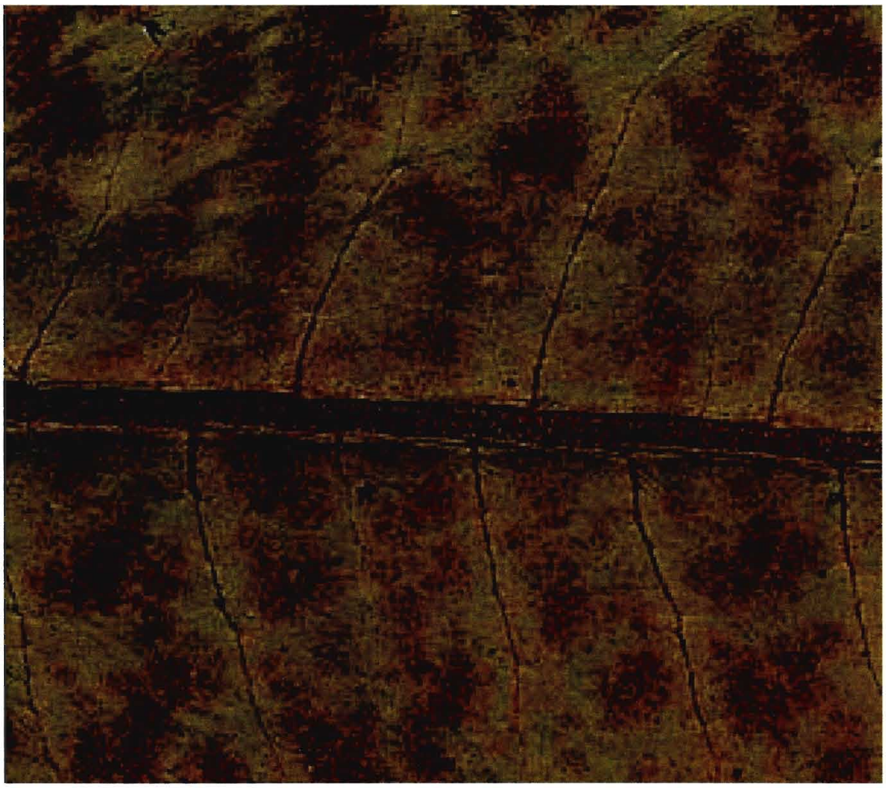
black halos and chlorosis



Blight injury from fungus



Heat stress



Low nutrients


Monarch mechanical damage


Tussock moths with mechanical damage

Ozone-induced injury from Kohut, 2005:

Stippling spreading dogbane (*Apocynum androsaemifolium*)

Chlorosis on Spreading dogbane (*Apocynum androsaemifolium*)


Bifacial Necrosis on aspen (*Populus tremuloides*).

Appendix III: Data as Entered into SAS version 9.1
ANOVA:

Legend for Appendix III:
SITE: B=Brantford, L=London, G=Grand Bend, S=Sarnia, E=Essex, C=Chatham, P=Port Stanley, SM=Simcoe
TIME/SURVEY: 1=August 06-08, 2=August 19-21, 3=August 26-28, 4=September 04-05, 5=September 10-11
INCIDENCE OF LEAVES:Incidence of leaves damaged per plant
INCIDENCE OF PLANTS:Incidence of plants damaged per site
TOTAL O3/TOZONE: Incidence-plants x average Horsfall-Barratt value per plant
SUM65: sum of ozone reading (ppb) each hour over 65 ppb
CUMO3=Cumulative O3:sum all O_3 values over zero
H-B:Average Horsfall-Barratt Value (0-5 Scale

ID	SITE	TIME	SUM65	CUM03	Incidence of leaves	Incidence of plants	Total O3	H-B	Precipitation (mm)	Temp (C°)
1	B	1	7921	60567	0	0	0	0	74.6	24
1	B	2	7921	68788	0	0	0	0	87.6	20.3
1	B	3	8055	73725	1.94	5.33	0.85	1.475	112.8	21
1	B	4	8804	79536	7.21	33.33	0.32	1.336	112.8	19.8
1	B	5	10072	83880	18.84	46.67	0.54	1.31	129.6	20.4
2	L	1	5050	59082	0	0	0	0	109.3	23.3
2	L	2	5050	66935	0	0	0	0	131.5	19
2	L	3	5050	71637	1.39	10.67	0.14	1.25	157.4	20.4
2	L	4	5684	77977	6.54	36	0.14	1.17	160.2	19.7
2	L	5	6225	82277	9.32	46.67	0.22	1.15	186.4	19.9
3	G	1	9650	60873	0	0	0	0	140.6	23.6
3	G	2	9650	68117	0	0	0	0	147.6	119.4
3	G	3	9650	72809	3.23	16	0.27	1.18	172.2	20.8
3	G	4	10786	79112	18.73	37.33	0.69	1.53	172.2	20.6
3	G	5	11286	83471	36.47	53.33	1.15	1.69	186.8	20.9
4	S	1	7702	59876	0	0	0	0	155.8	23.7
4	S	2	8002	67573	0	0	0	0	197.6	19.63
4	S	3	8070	72268	0.42	4	0.20	1.87	217.5	21
4	S	4	9240	78653	2.31	18.67	0.24	1.5	225.2	19.9
4	S	5	9240	82670	5.24	36	0.25	1.27	239.5	20.6
5	E	1	8076	62027	0	0	0	0	140.9	24.8
5	E	2	8666	71085	0	25.33	0.21	1	220.9	20.7
5	E	3	8733	76588	15.14	36	0.55	1.25	261.9	21.5
5	E	4	9628	83232	36.10	60	0.87	1.37	262.5	20.5
5	E	5	9628	86741	57.39	89.33	0.88	1.32	305.7	21.1
6	C	1	9692	67274	0	0	0	0	101.6	25.1
6	C	2	9983	76538	5.24	5.33	0.25	1	153.6	21.9
6	C	3	10123	82024	14.33	26.67	1.18	2.12	193.8	22.8
6	C	4	11883	89550	20.45	29.33	1.91	2.51	193.8	22.2
6	C	5	12518	94403	23.43	36	1.84	2.78	212.7	21.8
7	P	1	14439	69289	0	0	0	0	87.7	23.25
7	P	2	14572	78943	1.01	0	0	0	111.8	19.6
7	P	3	14934	84912	7.14	18.67	0.64	1.31	140.5	20.8
7	P	4	15557	91269	15.93	32	0.92	1.53	142.6	20.3

7	P	5	16731	95251	41.92	68	1.28	1.79	164.2	20.2
8	SM	1	9226	68597	0	0	0	0	121.9	22.4
8	SM	2	9432	77847	0	0	0	0	171.9	18.7
8	SM	3	9649	83705	21.16	62.67	0.52	1.6	200.6	20.5
8	SM	4	10439	90929	35.62	70.67	0.92	1.4	201.4	19.2
8	SM	5	11891	96226	37.52	76	0.75	1.47	239.2	20.6

MANOVA:

SITE	ID	Incidence of leaves SURVEY 1	SURVEY 2	SURVEY 3	SURVEY 4	SURVEY 5	Incidence of plants SURVEY 1	SURVEY 2
B	1	0	0	1.936041	7.211685	18.83929	0	0
L	2	0	0	1.397578	6.539206	9.316208	0	0
G	3	0	0	3.231791	18.73222	36.47264	0	0
S	4	0	0	0.417508	2.305336	5.240656	0	0
E	5	0	0	15.1356	36.10076	57.3948	0	25.33333
C	6	0	5.244977	14.33508	20.44757	23.42577	0	5.333333
P	7	0	1.010476	7.140522	15.93432	41.92279	0	0
M	8	0	0	21.1642	35.62421	37.51904	0	0

SURVEY 3	SURVEY 4	SURVEY 5	TOZONE SURVEY 1	SURVEY 2	SURVEY 3	SURVEY 4	SURVEY 5	SUM65 SURVEY 1	SURVEY 2
5.333333	33.33333	46.66667	0	0	0.850324	0.319316	0.543432	7921	7921
10.66667	36	46.66667	0	0	0.142387	0.141495	0.217405	5050	5050
16	37.33333	53.33333	0	0	0.266769	0.696531	1.150673	9650	9650
4	18.66667	36	0	0	0.198316	0.235644	0.248386	7702	8002
36	60	89.33333	0	0.207039	0.549405	0.868029	0.881859	8076	8666
26.66667	29.33333	36	0	0.252619	1.181889	1.914835	1.840214	9692	9983
18.66667	32	68	0	0	0.635516	0.919174	1.280835	14439	14572
62.66667	70.66667	76	0	0	0.524556	0.919174	0.744767	9226	9432

SURVEY 3	SURVEY 4	SURVEY 5	CUM03 SURVEY 1	SURVEY 2	SURVEY 3	SURVEY 4	SURVEY 5	Precipitation (mm) SURVEY 1	SURVEY 2
8055	8804	10072	60567	68788	73725	79536	83880	74.6	87.6
5050	5684	6225	59082	66935	71637	77977	82277	109.3	131.5
9650	10786	11286	60873	68117	72809	79112	83471	140.6	147.6
8070	9240	9240	59876	67573	72268	78653	82670	155.8	197.6
8733	9628	9628	62027	71085	76588	83232	86741	140.9	220.9
10123	11883	12518	67274	76538	82024	89550	94403	101.6	153.6
14934	15557	16731	69289	78943	84912	91269	95251	87.7	111.8
9649	10439	11891	68597	77847	83705	90929	96226	121.9	171.9

SURVEY 3	SURVEY 4	SURVEY 5
112.8	112.8	129.6
157.4	160.2	186.4
172.2	172.2	186.8
217.5	225.2	239.5
261.9	262.5	305.7
193.8	193.8	212.7
140.5	142.6	164.2
200.6	201.4	239.2

Appendix IV: Results of Statistical Analysis (using SAS version 9.1)

Legend for Appendix IV:
TIME/SURVEY: 1=August 06-08, 2=August 19-21, 3=August 26-28, 4=September 04-05, 5=September 10-11
INCIDENCE OF LEAVES:Incidence of leaves damaged per plant
INCIDENCE OF PLANTS:Incidence of plants damaged per site
TOTAL O3/TOZONE: Incidence-plants x average Horsfall-Barratt value per plant
SUM65: sum of ozone reading (ppb) each hour over 65 ppb
CUMO3=Cumulative O3:sum all O_3 values over zero

ANOVA Outputs:

Results from Table 5.4

Type 3 Tests of Fixed Effects: PLANTS AND SUM65

Effect	Num DF	Den DF	Chi-Square	F Value	Pr > ChiSq	Pr > F
TIME	1	28	0.00	0.00	0.9836	0.9838
SUM65	1	28	17.37	17.37	<.0001	0.0003
PREIP	1	28	5.32	5.32	0.0211	0.0287
TIME*SUM65	1	28	13.72	13.72	0.0002	0.0009

Type 3 Tests of Fixed Effects: LEAVES AND SUM65

Effect	Num DF	Den DF	Chi-Square	F Value	Pr > ChiSq	Pr > F
TIME	1	28	4.45	4.45	0.0350	0.0441
SUM65	1	28	16.49	16.49	<.0001	0.0004
PREIP	1	28	7.41	7.41	0.0065	0.0111
TIME*SUM65	1	28	26.98	26.98	<.0001	<.0001

Type 3 Tests of Fixed Effects: TOZONE AND SUM65

Effect	Num DF	Den DF	Chi-Square	F Value	Pr > ChiSq	Pr > F
TIME	1	28	2.41	2.41	0.1207	0.1320
SUM65	1	28	4.28	4.28	0.0385	0.0479
PREIP	1	28	1.33	1.33	0.2496	0.2593
TIME*SUM65	1	28	20.41	20.41	<.0001	0.0001

0002

Results from Table 5.5:

Type 3 Tests of Fixed Effects: LEAVES AND CUM03						
Effect	Num DF	Den DF	Chi-Square	F Value	Pr > ChiSq	Pr > F
TIME	1	28	16.31	16.31	<.0001	0.0004
CUM03	1	28	2.69	2.69	0.1010	0.1122
PREIP	1	28	7.38	7.38	0.0066	0.0112
TIME*CUM03	1	28	29.34	29.34	<.0001	<.0001

Type 3 Tests of Fixed Effects: PLANTS AND CUM03						
Effect	Num DF	Den DF	Chi-Square	F Value	Pr > ChiSq	Pr > F
TIME	1	28	6.39	6.39	0.0115	0.0174
CUM03	1	28	7.50	7.50	0.0062	0.0106
PREIP	1	28	8.40	8.40	0.0038	0.0072
TIME*CUM03	1	28	18.36	18.36	<.0001	0.0002

Type 3 Tests of Fixed Effects: TOTAL O3 AND CUMO3						
Effect	Num DF	Den DF	Chi-Square	F Value	Pr > ChiSq	Pr > F
TIME	1	28	10.85	10.85	0.0010	0.0027
CUM03	1	28	0.36	0.36	0.5495	0.5543
PREIP	1	28	0.16	0.16	0.6899	0.6930
TIME*CUM03	1	28	19.51	19.51	<.0001	0.0001

Results from Table 5.6:
Incidence of Leaves Damaged:

Source	DF	Type III SS	Mean Square	F Value	Pr > F
ID	7	766.7251579	109.5321654	1.34	0.2684
PREIP	1	63.1865209	63.1865209	0.77	0.3868
TEMP	1	27.5395483	27.5395483	0.34	0.5663
CUM03	1	342.1415914	342.1415914	4.18	0.0500

Incidence of Plants Damaged:

Source	DF	Type III SS	Mean Square	F Value	Pr > F
ID	7	2933.246440	419.035206	2.75	0.0255
PREIP	1	96.045773	96.045773	0.63	0.4336
TEMP	1	76.240979	76.240979	0.50	0.4849
CUM03	1	1809.798338	1809.798338	11.88	0.0018

Total Ozone:

Source	DF	Type III SS	Mean Square	F Value	Pr > F
ID	7	1.60926267	0.22989467	2.73	0.0265
PREIP	1	0.00939387	0.00939387	0.11	0.7409
TEMP	1	0.04994860	0.04994860	0.59	0.4477
CUM03	1	0.60166291	0.60166291	7.14	0.0123

MANOVA:
Combine all three dependent variables together and test for site effect
SUM65:

MANOVA Test Criteria and F Approximations for the Hypothesis of No Overall Site Effect S=3 M=1.5 N=12.5					
Statistic	Value	F Value	Num DF	Den DF	Pr > F
Wilks' Lambda	0.13664824	3.72	21	78.08	<.0001

CUMO3:

MANOVA Test Criteria and F Approximations for the Hypothesis of No Overall ID Effect H = Type III SSCP Matrix for ID E = Error SSCP Matrix S=3 M=1.5 N=12.5					
Statistic	Value	F Value	Num DF	Den DF	Pr > F
Wilks' Lambda	0.29353141	1.98	21	78.08	0.0162

<u>Now Test for Site effect individually, using ANOVA, no repeated statements used</u>
Results from Table 5.6:
Leaves Damaged per Plant

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Site	7	2145.701558	306.528794	4.38	0.0020
SUM65	1	686.362357	686.362357	9.81	0.0039
PREIP	1	440.514076	440.514076	6.30	0.0179
TEMP	1	19.431525	19.431525	0.28	0.6022

Plants Damaged per Site

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Site	7	8359.076728	1194.153818	7.87	<.0001
SUM65	1	1827.162440	1827.162440	12.04	0.0016
PREIP	1	2260.855911	2260.855911	14.90	0.0006
TEMP	1	66.886699	66.886699	0.44	0.5119

Total Ozone

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Site	7	3.63477695	0.51925385	6.56	0.0001
SUM65	1	0.75240946	0.75240946	9.51	0.0045
PREIP	1	0.44516413	0.44516413	5.63	0.0245
TEMP	1	0.04261508	0.04261508	0.54	0.4689

MANOVA:
Results from Section 5.4.2:
Leaves Damaged per Plant

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	1798.151065	256.878724	1.22	0.3222
Error	32	6753.788952	211.055905		
Corrected Total	39	8551.940016			

Plants Damaged per Site

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	4503.77778	643.39683	0.95	0.4826
Error	32	21653.33333	676.66667		
Corrected Total	39	26157.11111			

Total Ozone

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	2.97331619	0.42475946	1.85	0.1124
Error	32	7.36568911	0.23017778		
Corrected Total	39	10.33900530			

Appendix V: Table of *F*-Critical Values (p=0.05)

df2\df1	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	22	24	26	28	30	35	40	45	50	60	70	80	100	200	500	1000	>1000	df1/df2
3	10.13	9.55	9.28	9.12	9.01	8.94	8.89	8.85	8.81	8.79	8.76	8.74	8.73	8.71	8.70	8.69	8.68	8.67	8.67	8.66	8.65	8.64	8.63	8.62	8.62	8.60	8.59	8.59	8.58	8.57	8.57	8.56	8.55	8.54	8.53	8.53	8.54	3
4	7.71	6.94	6.59	6.39	6.26	6.16	6.09	6.04	6.00	5.96	5.94	5.91	5.89	5.87	5.86	5.84	5.83	5.82	5.81	5.80	5.79	5.77	5.76	5.75	5.75	5.73	5.72	5.71	5.70	5.69	5.68	5.67	5.66	5.65	5.64	5.63	5.63	4
5	6.61	5.79	5.41	5.19	5.05	4.95	4.88	4.82	4.77	4.74	4.70	4.68	4.66	4.64	4.62	4.60	4.59	4.58	4.57	4.56	4.54	4.53	4.52	4.50	4.50	4.48	4.46	4.45	4.44	4.43	4.42	4.42	4.41	4.39	4.37	4.37	4.36	5
6	5.99	5.14	4.76	4.53	4.39	4.28	4.21	4.15	4.10	4.06	4.03	4.00	3.98	3.96	3.94	3.92	3.91	3.90	3.88	3.87	3.86	3.84	3.83	3.82	3.81	3.79	3.77	3.76	3.75	3.74	3.73	3.72	3.71	3.69	3.68	3.67	3.67	6
7	5.59	4.74	4.35	4.12	3.97	3.87	3.79	3.73	3.68	3.64	3.60	3.57	3.55	3.53	3.51	3.49	3.48	3.47	3.46	3.44	3.43	3.41	3.40	3.39	3.38	3.36	3.34	3.33	3.32	3.30	3.29	3.29	3.27	3.25	3.24	3.23	3.23	7
8	5.32	4.46	4.07	3.84	3.69	3.58	3.50	3.44	3.39	3.35	3.31	3.28	3.26	3.24	3.22	3.20	3.19	3.17	3.16	3.15	3.13	3.12	3.10	3.09	3.08	3.06	3.04	3.03	3.02	3.01	2.99	2.99	2.97	2.95	2.94	2.93	2.93	8
9	5.12	4.26	3.86	3.63	3.48	3.37	3.29	3.23	3.18	3.14	3.10	3.07	3.05	3.03	3.01	2.99	2.97	2.96	2.95	2.94	2.92	2.90	2.89	2.87	2.86	2.84	2.83	2.81	2.80	2.79	2.78	2.77	2.76	2.73	2.72	2.71	2.71	9
10	4.96	4.10	3.71	3.48	3.33	3.22	3.14	3.07	3.02	2.98	2.94	2.91	2.89	2.86	2.85	2.83	2.81	2.80	2.79	2.77	2.75	2.74	2.72	2.71	2.70	2.68	2.66	2.65	2.64	2.62	2.61	2.60	2.59	2.56	2.55	2.54	2.54	10
11	4.84	3.98	3.59	3.36	3.20	3.09	3.01	2.95	2.90	2.85	2.82	2.79	2.76	2.74	2.72	2.70	2.69	2.67	2.66	2.65	2.63	2.61	2.59	2.58	2.57	2.55	2.53	2.52	2.51	2.49	2.48	2.47	2.46	2.43	2.42	2.41	2.41	11
12	4.75	3.89	3.49	3.26	3.11	3.00	2.91	2.85	2.80	2.75	2.72	2.69	2.66	2.64	2.62	2.60	2.58	2.57	2.56	2.54	2.52	2.51	2.49	2.48	2.47	2.44	2.43	2.41	2.40	2.38	2.37	2.36	2.35	2.32	2.31	2.30	2.30	12
13	4.67	3.81	3.41	3.18	3.03	2.92	2.83	2.77	2.71	2.67	2.63	2.60	2.58	2.55	2.53	2.51	2.50	2.48	2.47	2.46	2.44	2.42	2.41	2.39	2.38	2.36	2.34	2.33	2.31	2.30	2.28	2.27	2.26	2.23	2.22	2.21	2.21	13
14	4.60	3.74	3.34	3.11	2.96	2.85	2.76	2.70	2.65	2.60	2.57	2.53	2.51	2.48	2.46	2.44	2.43	2.41	2.40	2.39	2.37	2.35	2.33	2.32	2.31	2.28	2.27	2.25	2.24	2.22	2.21	2.20	2.19	2.16	2.14	2.14	2.13	14
15	4.54	3.68	3.29	3.06	2.90	2.79	2.71	2.64	2.59	2.54	2.51	2.48	2.45	2.42	2.40	2.38	2.37	2.35	2.34	2.33	2.31	2.29	2.27	2.26	2.25	2.22	2.20	2.19	2.18	2.16	2.15	2.14	2.12	2.10	2.08	2.07	2.07	15
16	4.49	3.63	3.24	3.01	2.85	2.74	2.66	2.59	2.54	2.49	2.46	2.42	2.40	2.37	2.35	2.33	2.32	2.30	2.29	2.28	2.25	2.24	2.22	2.21	2.19	2.17	2.15	2.14	2.12	2.11	2.09	2.08	2.07	2.04	2.02	2.02	2.01	16
17	4.45	3.59	3.20	2.96	2.81	2.70	2.61	2.55	2.49	2.45	2.41	2.38	2.35	2.33	2.31	2.29	2.27	2.26	2.24	2.23	2.21	2.19	2.17	2.16	2.15	2.12	2.10	2.09	2.08	2.06	2.05	2.03	2.02	1.99	1.97	1.97	1.96	17
18	4.41	3.55	3.16	2.93	2.77	2.66	2.58	2.51	2.46	2.41	2.37	2.34	2.31	2.29	2.27	2.25	2.23	2.22	2.20	2.19	2.17	2.15	2.13	2.12	2.11	2.08	2.06	2.05	2.04	2.02	2.00	1.99	1.98	1.95	1.93	1.92	1.92	18
19	4.38	3.52	3.13	2.90	2.74	2.63	2.54	2.48	2.42	2.38	2.34	2.31	2.28	2.26	2.23	2.21	2.20	2.18	2.17	2.16	2.13	2.11	2.10	2.08	2.07	2.05	2.03	2.01	2.00	1.98	1.97	1.96	1.94	1.91	1.89	1.88	1.88	19
20	4.35	3.49	3.10	2.87	2.71	2.60	2.51	2.45	2.39	2.35	2.31	2.28	2.25	2.23	2.20	2.18	2.17	2.15	2.14	2.12	2.10	2.08	2.07	2.05	2.04	2.01	1.99	1.98	1.97	1.95	1.93	1.92	1.91	1.88	1.86	1.85	1.84	20
22	4.30	3.44	3.05	2.82	2.66	2.55	2.46	2.40	2.34	2.30	2.26	2.23	2.20	2.17	2.15	2.13	2.11	2.10	2.08	2.07	2.05	2.03	2.01	2.00	1.98	1.96	1.94	1.92	1.91	1.89	1.88	1.86	1.85	1.82	1.80	1.79	1.78	22
24	4.26	3.40	3.01	2.78	2.62	2.51	2.42	2.36	2.30	2.25	2.22	2.18	2.15	2.13	2.11	2.09	2.07	2.05	2.04	2.03	2.00	1.98	1.97	1.95	1.94	1.91	1.89	1.88	1.86	1.84	1.83	1.82	1.80	1.77	1.75	1.74	1.73	24
26	4.23	3.37	2.98	2.74	2.59	2.47	2.39	2.32	2.27	2.22	2.18	2.15	2.12	2.09	2.07	2.05	2.03	2.02	2.00	1.99	1.97	1.95	1.93	1.91	1.90	1.87	1.85	1.84	1.82	1.80	1.79	1.78	1.76	1.73	1.71	1.70	1.69	26
28	4.20	3.34	2.95	2.71	2.56	2.45	2.36	2.29	2.24	2.19	2.15	2.12	2.09	2.06	2.04	2.02	2.00	1.99	1.97	1.96	1.93	1.91	1.90	1.88	1.87	1.84	1.82	1.80	1.79	1.77	1.75	1.74	1.73	1.69	1.67	1.66	1.66	28
30	4.17	3.32	2.92	2.69	2.53	2.42	2.33	2.27	2.21	2.16	2.13	2.09	2.06	2.04	2.01	1.99	1.98	1.96	1.95	1.93	1.91	1.89	1.87	1.85	1.84	1.81	1.79	1.77	1.76	1.74	1.72	1.71	1.70	1.66	1.64	1.63	1.62	30
35	4.12	3.27	2.87	2.64	2.49	2.37	2.29	2.22	2.16	2.11	2.08	2.04	2.01	1.99	1.96	1.94	1.92	1.91	1.89	1.88	1.85	1.83	1.82	1.80	1.79	1.76	1.74	1.72	1.70	1.68	1.66	1.65	1.63	1.60	1.57	1.57	1.56	35
40	4.08	3.23	2.84	2.61	2.45	2.34	2.25	2.18	2.12	2.08	2.04	2.00	1.97	1.95	1.92	1.90	1.89	1.87	1.85	1.84	1.81	1.79	1.77	1.76	1.74	1.72	1.69	1.67	1.66	1.64	1.62	1.61	1.59	1.55	1.53	1.52	1.51	40
45	4.06	3.20	2.81	2.58	2.42	2.31	2.22	2.15	2.10	2.05	2.01	1.97	1.94	1.92	1.89	1.87	1.86	1.84	1.82	1.81	1.78	1.76	1.74	1.73	1.71	1.68	1.66	1.64	1.63	1.60	1.59	1.57	1.55	1.51	1.49	1.48	1.47	45
50	4.03	3.18	2.79	2.56	2.40	2.29	2.20	2.13	2.07	2.03	1.99	1.95	1.92	1.89	1.87	1.85	1.83	1.81	1.80	1.78	1.76	1.74	1.72	1.70	1.69	1.66	1.63	1.61	1.60	1.58	1.56	1.54	1.52	1.48	1.46	1.45	1.44	50
60	4.00	3.15	2.76	2.53	2.37	2.25	2.17	2.10	2.04	1.99	1.95	1.92	1.89	1.86	1.84	1.82	1.80	1.78	1.76	1.75	1.72	1.70	1.68	1.66	1.65	1.62	1.59	1.57	1.56	1.53	1.52	1.50	1.48	1.44	1.41	1.40	1.39	60
70	3.98	3.13	2.74	2.50	2.35	2.23	2.14	2.07	2.02	1.97	1.93	1.89	1.86	1.84	1.81	1.79	1.77	1.75	1.74	1.72	1.70	1.67	1.65	1.64	1.62	1.59	1.57	1.55	1.53	1.50	1.49	1.47	1.45	1.40	1.37	1.36	1.35	70
80	3.96	3.11	2.72	2.49	2.33	2.21	2.13	2.06	2.00	1.95	1.91	1.88	1.84	1.82	1.79	1.77	1.75	1.73	1.72	1.70	1.68	1.65	1.63	1.62	1.60	1.57	1.54	1.52	1.51	1.48	1.46	1.45	1.43	1.38	1.35	1.34	1.33	80
100	3.94	3.09	2.70	2.46	2.31	2.19	2.10	2.03	1.97	1.93	1.89	1.85	1.82	1.79	1.77	1.75	1.73	1.71	1.69	1.68	1.65	1.63	1.61	1.59	1.57	1.54	1.52	1.49	1.48	1.45	1.43	1.41	1.39	1.34	1.31	1.30	1.28	100
200	3.89	3.04	2.65	2.42	2.26	2.14	2.06	1.98	1.93	1.88	1.84	1.80	1.77	1.74	1.72	1.69	1.67	1.66	1.64	1.62	1.60	1.57	1.55	1.53	1.52	1.48	1.46	1.43	1.41	1.39	1.36	1.35	1.32	1.26	1.22	1.21	1.19	200

Appendix VI: Raw Data Collected During Milkweed Surveys

August 06-08 2007 - Brantford:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	85	17	9	0	0	0	0
2	110	20	11	0	4	0	0
3	74	14	8	0	0	0	0
4	99	11	6	0	3	0	0
5	82	13	7	0	3	0	0
6	115	18	9	0	4	0	0
7	100	14	7	0	1	0	0
8	127	14	8	0	4	0	0
9	78	7	4	0	3	0	0
10	73	6	4	0	0	0	0
11	79	6	4	0	0	0	0
12	115	12	7	0	3	0	0
13	112	17	9	0	6	0	0
14	120	15	8	0	4	0	0
15	99	16	8	0	1	0	0
16	120	11	6	0	4	0	0
17	134	13	7	0	3	0	0
18	114	8	5	0	3	0	0
19	131	14	7	0	11	0	0
20	120	5	4	0	3	0	0
21	100	13	7	0	4	0	0
22	98	19	10	0	0	0	0
23	77	15	8	0	0	0	0
24	90	12	7	0	0	0	0
25	95	20	11	0	0	0	0
26	94	12	7	0	0	0	0
27	99	18	7	0	0	0	0
28	103	22	12	0	0	0	0
29	108	25	13	0	1	0	0
30	90	18	10	0	0	0	0
31	78	12	7	0	0	0	0
32	110	23	12	0	0	0	0
33	96	13	8	0	2	0	0
34	84	14	9	0	0	0	0
35	71	18	9	0	0	0	0
36	90	21	11	0	0	0	0
37	77	18	10	0	0	0	0
38	81	13	9	0	0	0	0
39	113	17	9	0	10	0	0
40	90	12	6	0	3	0	0
41	88	13	7	0	1	0	0
42	89	15	7	0	0	0	0

43	97	22	11	0	1	0	0
44	70	15	9	0	0	0	0
45	125	25	13	0	4	0	0
46	109	10	5	0	0	0	0
47	150	20	9	0	9	0	0
48	98	15	8	0	0	0	0
49	70	19	10	0	0	0	0
50	79	19	10	0	0	0	0
51	71	13	7	0	1	0	0
52	79	19	10	0	0	0	0
53	102	13	9	0	3	0	0
54	110	23	12	0	2	0	0
55	70	16	9	0	0	0	0
56	105	18	11	0	5	0	0
57	74	11	7	0	3	0	0
58	90	13	8	0	1	0	0
59	132	21	12	0	4	0	0
60	66	12	7	0	0	0	0
61	86	14	8	0	1	0	0
62	100	15	8	0	4	0	0
63	68	9	5	0	0	0	0
64	100	12	8	0	2	0	0
65	103	10	8	0	0	0	0
66	96	7	5	0	1	0	0
67	108	12	8	0	3	0	0
68	96	10	5	0	1	0	0
69	87	11	8	0	2	0	0
70	92	13	7	0	3	0	0
71	84	17	9	0	0	0	0
72	94	12	7	0	2	0	0
73	95	17	9	0	3	0	0
74	100	8	5	0	0	0	0
75	118	11	6	0	3	0	0

August 06-08 2007 – London:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	75	15	8	0	0	0	0
2	68	14	7	0	0	0	0
3	80	17	9	0	0	0	0
4	104	15	9	0	6	0	0
5	85	24	12	0	0	0	0
6	98	12	6	0	5	0	0
7	100	20	10	0	2	0	0
8	88	20	11	0	0	0	0
9	80	18	9	0	1	0	0
10	83	20	10	0	0	0	0

11	76	15	8	0	0	0	0
12	94	20	10	0	0	0	0
13	81	14	8	0	0	0	0
14	98	14	8	0	0	0	0
15	81	14	8	0	0	0	0
16	88	15	8	0	1	0	0
17	99	17	9	0	0	0	0
18	113	22	11	0	2	0	0
19	109	25	13	0	1	0	0
20	120	14	9	0	8	0	0
21	107	21	11	0	2	0	0
22	81	16	9	0	0	0	0
23	91	19	10	0	0	0	0
24	93	24	12	0	0	0	0
25	81	14	8	0	0	0	0
26	100	27	14	0	0	0	0
27	137	27	14	0	4	0	0
28	94	20	11	0	0	0	0
29	98	19	12	0	0	0	0
30	86	13	8	0	0	0	0
31	85	16	8	0	0	0	0
32	90	17	9	0	0	0	0
33	138	27	15	0	3	0	0
34	118	30	15	0	0	0	0
35	130	26	13	0	0	0	0
36	103	20	11	0	0	0	0
37	95	17	12	0	0	0	0
38	156	31	16	0	7	0	0
39	111	22	11	0	2	0	0
40	126	25	13	0	5	0	0
41	112	26	13	0	0	0	0
42	112	23	12	0	2	0	0
43	118	22	11	0	4	0	0
44	132	26	13	0	8	0	0
45	104	19	10	0	2	0	0
46	114	21	11	0	4	0	0
47	111	21	11	0	2	0	0
48	125	15	8	0	8	0	0
49	88	15	8	0	0	0	0
50	100	23	12	0	0	0	0
51	115	29	15	0	0	0	0
52	80	20	11	0	0	0	0
53	104	21	11	0	0	0	0
54	98	21	11	0	0	0	0
55	106	22	12	0	0	0	0
56	124	27	14	0	0	0	0
57	110	23	12	0	0	0	0
58	98	24	13	0	0	0	0

59	87	19	10	0	0	0	0
60	89	19	10	0	0	0	0
61	86	21	11	0	0	0	0
62	101	23	12	0	0	0	0
63	96	17	9	0	0	0	0
64	118	18	9	0	0	0	0
65	100	14	8	0	0	0	0
66	125	15	9	0	4	0	0
67	138	21	11	0	5	0	0
68	121	17	10	0	4	0	0
69	89	15	9	0	0	0	0
70	94	17	9	0	2	0	0
71	93	15	10	0	3	0	0
72	97	16	12	0	0	0	0
73	80	13	9	0	1	0	0
74	86	15	10	0	1	0	0
75	110	25	12	0	6	0	0

August 06-08 2007 – Grand Bend:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	86	21	11	0	1	0	0
2	74	12	6	0	0	0	0
3	114	19	10	0	6	0	0
4	134	28	14	0	1	0	0
5	140	26	14	0	5	0	0
6	113	17	9	0	4	0	0
7	105	13	7	0	3	0	0
8	96	17	9	0	5	0	0
9	92	12	7	0	0	0	0
10	74	15	8	0	5	0	0
11	127	28	14	0	2	0	0
12	139	26	14	0	5	0	0
13	121	19	10	0	2	0	0
14	133	23	12	0	4	0	0
15	118	25	14	0	4	0	0
16	82	20	11	0	0	0	0
17	99	20	11	0	1	0	0
18	105	18	11	0	1	0	0
19	91	15	9	0	4	0	0
20	95	21	13	0	2	0	0
21	99	23	13	0	7	0	0
22	84	20	10	0	3	0	0
23	86	15	8	0	1	0	0
24	99	19	10	0	4	0	0
25	108	24	12	0	7	0	0
26	103	23	12	0	10	0	0

27	104	18	11	0	8	0	0
28	98	20	12	0	6	0	0
29	94	20	11	0	5	0	0
30	124	17	12	0	6	0	0
31	94	16	10	0	4	0	0
32	93	17	10	0	3	0	0
33	105	19	11	0	3	0	0
34	115	20	12	0	3	0	0
35	115	14	8	0	2	0	0
36	130	21	12	0	0	0	0
37	114	25	13	0	1	0	0
38	67	15	8	0	0	0	0
39	118	20	10	0	1	0	0
40	127	20	11	0	2	0	0
41	138	20	11	0	0	0	0
42	136	10	6	0	7	0	0
43	134	20	10	0	3	0	0
44	134	21	11	0	5	0	0
45	114	19	10	0	1	0	0
46	115	20	10	0	0	0	0
47	120	19	10	0	2	0	0
48	122	22	11	0	0	0	0
49	99	21	11	0	0	0	0
50	120	22	11	0	0	0	0
51	113	20	10	0	0	0	0
52	125	24	9	0	2	0	0
53	130	25	11	0	6	0	0
54	135	22	10	0	3	0	0
55	127	22	11	0	3	0	0
56	91	21	11	0	0	0	0
57	96	22	11	0	0	0	0
58	96	16	8	0	0	0	0
59	100	20	10	0	0	0	0
60	100	16	8	0	0	0	0
61	130	24	11	0	6	0	0
62	130	25	13	0	4	0	0
63	124	19	10	0	3	0	0
64	98	17	8	0	4	0	0
65	123	20	10	0	6	0	0
66	105	25	13	0	0	0	0
67	89	20	10	0	0	0	0
68	80	28	15	0	0	0	0
69	102	22	13	0	0	0	0
70	101	24	13	0	3	0	0
71	97	20	11	0	3	0	0
72	86	23	12	0	0	0	0
73	97	18	12	0	1	0	0
74	99	18	9	0	2	0	0

75	121	18	11	0	3	0	0

August 06-08 2007– Sarnia:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	105	24	12	0	4	0	0
2	99	22	11	0	4	0	0
3	73	17	9	0	0	0	0
4	74	22	12	0	0	0	0
5	74	21	11	0	0	0	0
6	89	18	10	0	4	0	0
7	88	18	9	0	0	0	0
8	87	20	10	0	0	0	0
9	97	13	7	0	3	0	0
10	110	17	9	0	9	0	0
11	112	19	10	0	5	0	0
12	98	24	12	0	0	0	0
13	114	17	10	0	7	0	0
14	102	17	9	0	4	0	0
15	117	19	10	0	4	0	0
16	113	17	9	0	6	0	0
17	100	20	11	0	4	0	0
18	93	21	11	0	0	0	0
19	70	19	10	0	0	0	0
20	79	18	9	0	0	0	0
21	82	17	9	0	0	0	0
22	74	16	8	0	0	0	0
23	81	16	8	0	0	0	0
24	98	14	8	0	3	0	0
25	85	18	9	0	0	0	0
26	71	15	8	0	0	0	0
27	82	14	7	0	0	0	0
28	81	15	9	0	0	0	0
29	80	14	7	0	0	0	0
30	90	12	6	0	1	0	0
31	103	15	9	0	1	0	0
32	76	16	8	0	0	0	0
33	75	14	7	0	0	0	0
34	74	14	7	0	0	0	0
35	104	22	11	0	3	0	0
36	99	20	11	0	2	0	0
37	74	17	9	0	0	0	0
38	100	17	9	0	1	0	0
39	96	17	9	0	0	0	0
40	94	23	12	0	1	0	0
41	89	18	10	0	3	0	0
42	74	18	10	0	1	0	0

43	81	24	12	0	2	0	0
44	98	26	13	0	6	0	0
45	110	24	12	0	5	0	0
46	86	23	12	0	2	0	0
47	83	25	13	0	0	0	0
48	97	20	10	0	4	0	0
49	98	19	10	0	4	0	0
50	96	16	8	0	2	0	0
51	132	23	12	0	10	0	0
52	116	27	14	0	3	0	0
53	126	29	15	0	6	0	0
54	86	24	12	0	0	0	0
55	82	19	10	0	0	0	0
56	93	21	11	0	2	0	0
57	89	21	11	0	1	0	0
58	73	17	9	0	0	0	0
59	74	19	10	0	0	0	0
60	73	18	10	0	0	0	0
61	120	30	15	0	1	0	0
62	79	16	8	0	0	0	0
63	91	32	16	0	0	0	0
64	67	18	9	0	0	0	0
65	107	28	14	0	4	0	0
66	94	24	12	0	0	0	0
67	99	26	13	0	0	0	0
68	100	22	11	0	5	0	0
69	75	24	12	0	1	0	0
70	110	25	13	0	6	0	0
71	99	26	13	0	4	0	0
72	90	18	9	0	4	0	0
73	61	16	8	0	0	0	0
74	64	18	9	0	0	0	0
75	66	19	10	0	0	0	0

August 06-08 2007– Essex:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	117	22	11	0	4	0	0
2	94	18	9	0	1	0	0
3	99	18	10	0	2	0	0
4	99	19	10	0	1	0	0
5	110	17	9	0	2	0	0
6	123	21	11	0	3	0	0
7	115	21	11	0	2	0	0
8	115	22	11	0	2	0	0
9	94	22	12	0	2	0	0
10	99	17	9	0	3	0	0

11	99	21	11	0	2	0	0
12	94	19	10	0	2	0	0
13	98	17	9	0	1	0	0
14	89	26	13	0	0	0	0
15	84	23	12	0	0	0	0
16	99	21	11	0	2	0	0
17	91	18	10	0	2	0	0
18	93	25	13	0	0	0	0
19	94	24	12	0	1	0	0
20	94	19	10	0	3	0	0
21	118	24	13	0	7	0	0
22	96	21	11	0	3	0	0
23	99	25	13	0	4	0	0
24	112	26	13	0	7	0	0
25	110	25	13	0	4	0	0
26	93	17	9	0	4	0	0
27	77	20	10	0	2	0	0
28	100	28	15	0	2	0	0
29	92	30	16	0	0	0	0
30	127	21	11	0	3	0	0
31	153	28	15	0	6	0	0
32	135	18	10	0	3	0	0
33	145	25	13	0	6	0	0
34	100	21	11	0	4	0	0
35	114	25	13	0	5	0	0
36	111	21	11	0	4	0	0
37	112	19	10	0	3	0	0
38	98	23	12	0	3	0	0
39	98	17	9	0	3	0	0
40	110	17	9	0	7	0	0
41	115	25	13	0	4	0	0
42	95	23	12	0	2	0	0
43	103	18	9	0	6	0	0
44	95	21	11	0	2	0	0
45	110	25	14	0	4	0	0
46	92	18	10	0	1	0	0
47	102	26	13	0	2	0	0
48	121	23	12	0	6	0	0
49	91	18	10	0	7	0	0
50	105	20	12	0	4	0	0
51	95	17	11	0	3	0	0
52	124	17	9	0	7	0	0
53	115	16	10	0	5	0	0
54	95	23	12	0	3	0	0
55	108	20	11	0	4	0	0
56	105	23	12	0	3	0	0
57	110	20	11	0	5	0	0
58	111	18	9	0	4	0	0

59	124	24	12	0	8	0	0
60	116	14	9	0	6	0	0
61	97	15	8	0	1	0	0
62	96	18	10	0	1	0	0
63	110	19	13	0	1	0	0
64	96	19	10	0	2	0	0
65	87	20	10	0	2	0	0
66	91	22	11	0	4	0	0
67	91	19	10	0	2	0	0
68	89	21	11	0	4	0	0
69	95	23	12	0	3	0	0
70	102	32	16	0	3	0	0
71	95	21	12	0	2	0	0
72	96	21	12	0	2	0	0
73	115	27	15	0	4	0	0
74	118	19	11	0	2	0	0
75	111	20	11	0	2	0	0

August 06-08 2007– Chatham:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	93	19	11	0	0	0	0
2	90	10	5	0	0	0	0
3	149	28	15	0	0	0	0
4	69	17	9	0	0	0	0
5	68	18	9	0	0	0	0
6	112	24	12	0	3	0	0
7	141	26	14	0	4	0	0
8	119	24	12	0	6	0	0
9	117	22	11	0	0	0	0
10	68	21	11	0	0	0	0
11	100	23	12	0	0	0	0
12	100	17	10	0	0	0	0
13	95	20	10	0	0	0	0
14	100	20	10	0	0	0	0
15	98	16	8	0	0	0	0
16	88	21	11	0	0	0	0
17	98	22	12	0	0	0	0
18	99	24	13	0	0	0	0
19	78	16	8	0	0	0	0
20	70	20	10	0	0	0	0
21	66	17	9	0	0	0	0
22	72	17	9	0	0	0	0
23	88	14	7	0	0	0	0
24	88	14	7	0	0	0	0
25	81	13	7	0	0	0	0

26	64	14	7	0	0	0	0
27	80	18	10	0	0	0	0
28	80	17	9	0	0	0	0
29	87	25	13	0	0	0	0
30	86	20	10	0	0	0	0
31	90	21	11	0	0	0	0
32	96	25	13	0	0	0	0
33	99	26	13	0	0	0	0
34	88	24	13	0	0	0	0
35	96	28	14	0	0	0	0
36	59	19	12	0	0	0	0
37	74	25	16	0	0	0	0
38	71	20	11	0	0	0	0
39	102	32	16	0	0	0	0
40	110	26	13	0	2	0	0
41	110	31	16	0	0	0	0
42	95	28	14	0	0	0	0
43	71	12	6	0	0	0	0
44	93	25	13	0	0	0	0
45	93	23	13	0	0	0	0
46	98	25	13	0	0	0	0
47	89	17	9	0	0	0	0
48	81	24	12	0	0	0	0
49	74	21	11	0	0	0	0
50	130	18	9	0	3	0	0
51	105	22	11	0	0	0	0
52	125	16	8	0	4	0	0
53	81	20	11	0	0	0	0
54	60	14	7	0	0	0	0
55	120	16	8	0	3	0	0
56	125	18	10	0	1	0	0
57	62	26	13	0	0	0	0
58	61	12	6	0	0	0	0
59	62	19	10	0	0	0	0
60	63	18	10	0	0	0	0
61	79	21	11	0	0	0	0
62	72	8	4	0	0	0	0
63	58	13	7	0	0	0	0
64	67	18	9	0	0	0	0
65	63	16	8	0	0	0	0
66	90	20	14	0	0	0	0
67	79	24	13	0	0	0	0
68	83	23	12	0	0	0	0
69	61	20	10	0	0	0	0
70	60	18	9	0	0	0	0
71	62	17	9	0	0	0	0
72	62	12	6	0	0	0	0
73	72	19	10	0	0	0	0

74	67	15	8	0	0	0	0
75	87	18	9	0	0	0	0

August 06-08 2007– Port Stanley:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	95	20	11	0	0	0	0
2	98	15	8	0	0	0	0
3	108	18	9	0	0	0	0
4	98	17	9	0	0	0	0
5	97	16	8	0	0	0	0
6	104	14	7	0	0	0	0
7	96	14	7	0	0	0	0
8	108	14	8	0	2	0	0
9	98	17	9	0	0	0	0
10	99	12	6	0	0	0	0
11	108	12	7	0	1	0	0
12	99	17	9	0	1	0	0
13	110	16	8	0	1	0	0
14	104	16	8	0	0	0	0
15	110	16	8	0	3	0	0
16	109	13	7	0	2	0	0
17	107	13	7	0	3	0	0
18	101	14	8	0	1	0	0
19	90	12	7	0	1	0	0
20	96	12	7	0	0	0	0
21	91	15	8	0	1	0	0
22	90	12	7	0	0	0	0
23	97	11	6	0	0	0	0
24	97	12	7	0	0	0	0
25	96	17	9	0	0	0	0
26	98	13	7	0	1	0	0
27	89	19	10	0	0	0	0
28	99	11	7	0	1	0	0
29	78	22	11	0	0	0	0
30	71	22	11	0	0	0	0
31	74	14	8	0	0	0	0
32	84	14	8	0	1	0	0
33	78	18	10	0	1	0	0
34	81	15	8	0	0	0	0
35	101	20	10	0	4	0	0
36	109	19	10	0	2	0	0
37	78	19	10	0	0	0	0
38	90	19	10	0	0	0	0
39	76	22	11	0	0	0	0
40	74	20	10	0	0	0	0
41	120	18	9	0	0	0	0

42	90	24	12	0	0	0	0
43	90	31	18	0	0	0	0
44	128	18	10	0	2	0	0
45	140	13	13	0	4	0	0
46	150	10	10	0	3	0	0
47	120	8	8	0	2	0	0
48	130	7	7	0	0	0	0
49	125	6	6	0	6	0	0
50	100	8	8	0	2	0	0
51	117	8	8	0	2	0	0
52	120	11	11	0	1	0	0
53	101	11	11	0	4	0	0
54	80	10	10	0	0	0	0
55	91	11	11	0	5	0	0
56	94	16	8	0	0	0	0
57	98	10	5	0	0	0	0
58	120	19	10	0	5	0	0
59	130	20	11	0	2	0	0
60	125	18	9	0	3	0	0
61	126	19	10	0	3	0	0
62	124	12	6	0	3	0	0
63	145	12	6	0	6	0	0
64	143	20	10	0	2	0	0
65	70	18	9	0	0	0	0
66	72	14	7	0	0	0	0
67	96	12	12	0	0	0	0
68	80	7	7	0	0	0	0
69	95	7	7	0	0	0	0
70	105	9	9	0	0	0	0
71	80	7	7	0	0	0	0
72	110	13	13	0	0	0	0
73	102	10	10	0	0	0	0
74	113	12	12	0	0	0	0
75	108	9	9	0	0	0	0

August 06-08 2007– Simcoe:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	80	19	10	0	0	0	0
2	108	24	12	0	0	0	0
3	95	22	11	0	1	0	0
4	94	20	10	0	0	0	0
5	106	18	10	0	4	0	0
6	103	18	9	0	4	0	0
7	105	20	10	0	4	0	0
8	130	20	9	0	8	0	0
9	140	20	10	0	9	0	0
10	139	19	9	0	5	0	0
11	138	19	9	0	3	0	0

12	118	18	9	0	5	0	0
13	75	15	8	0	0	0	0
14	95	19	10	0	5	0	0
15	99	19	10	0	6	0	0
16	99	23	12	0	3	0	0
17	125	20	11	0	8	0	0
18	124	22	11	0	11	0	0
19	135	20	9	0	6	0	0
20	102	17	8	0	9	0	0
21	67	16	8	0	0	0	0
22	78	18	9	0	0	0	0
23	88	16	9	0	0	0	0
24	80	17	9	0	0	0	0
25	91	20	11	0	5	0	0
26	110	21	11	0	0	0	0
27	110	21	11	0	7	0	0
28	99	22	11	0	9	0	0
29	86	14	8	0	3	0	0
30	103	19	10	0	11	0	0
31	73	19	9	0	0	0	0
32	121	19	10	0	10	0	0
33	123	21	11	0	9	0	0
34	120	21	12	0	11	0	0
35	117	23	13	0	13	0	0
36	108	17	11	0	6	0	0
37	84	17	10	0	6	0	0
38	99	22	11	0	5	0	0
39	111	24	13	0	10	0	0
40	90	19	10	0	4	0	0
41	92	13	9	0	7	0	0
42	81	18	9	0	0	0	0
43	90	20	10	0	5	0	0
44	117	17	9	0	6	0	0
45	121	23	13	0	9	0	0
46	122	29	15	0	11	0	0
47	97	19	10	0	0	0	0
48	90	21	11	0	0	0	0
49	98	18	9	0	0	0	0
50	99	26	13	0	0	0	0
51	86	22	11	0	0	0	0
52	115	21	11	0	0	0	0
53	116	24	12	0	0	0	0
54	89	24	12	0	0	0	0
55	76	22	11	0	0	0	0
56	86	22	11	0	0	0	0
57	69	20	10	0	0	0	0
58	79	20	10	0	0	0	0
59	90	22	11	0	0	0	0
60	84	21	11	0	4	0	0
61	66	22	11	0	0	0	0
62	75	24	12	0	0	0	0

63	62	20	10	0	0	0	0
64	83	24	12	0	0	0	0
65	64	18	9	0	0	0	0
66	61	19	10	0	0	0	0
67	71	23	12	0	0	0	0
68	64	20	10	0	0	0	0
69	77	24	12	0	0	0	0
70	72	20	10	0	0	0	0
71	66	22	11	0	0	0	0
72	68	13	7	0	0	0	0
73	76	24	12	0	0	0	0
74	76	19	10	0	0	0	0
75	60	22	11	0	0	0	0

August 19-21 2007– Brantford:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	82	16	8	0	0	0	0
2	58	15	8	0	0	0	0
3	55	12	7	0	0	0	0
4	115	15	9	0	3	0	0
5	100	10	6	0	3	0	0
6	76	12	6	0	0	0	0
7	83	12	6	0	3	0	0
8	111	12	7	0	4	0	0
9	114	17	9	0	4	0	0
10	80	6	4	0	2	0	0
11	75	4	3	0	0	0	0
12	125	15	8	0	6	0	0
13	117	12	6	0	3	0	0
14	133	9	7	0	3	0	0
15	135	10	6	0	4	0	0
16	135	13	7	0	7	0	0
17	130	3	3	0	3	0	0
18	112	0	0	0	2	0	0
19	69	7	6	0	0	0	0
20	99	18	9	0	0	0	0
21	69	12	7	0	0	0	0
22	112	18	9	0	0	0	0
23	99	18	10	0	0	0	0
24	114	24	12	0	1	0	0
25	100	17	9	0	0	0	0
26	84	18	9	0	0	0	0
27	73	11	6	0	0	0	0
28	66	6	4	0	0	0	0
29	115	20	11	0	0	0	0
30	98	9	6	0	2	0	0
31	82	17	9	0	0	0	0

32	80	15	8	0	0	0	0
33	83	13	9	0	0	0	0
34	75	19	10	0	0	0	0
35	86	20	11	0	0	0	0
36	71	18	10	0	0	0	0
37	90	10	7	0	0	0	0
38	115	16	9	0	1	0	0
39	90	12	6	0	0	0	0
40	87	17	8	0	3	0	0
41	85	16	8	0	1	0	0
42	70	16	8	0	1	0	0
43	90	20	10	0	0	0	0
44	120	25	13	0	4	0	0
45	56	19	10	0	0	0	0
46	96	10	5	0	0	0	0
47	107	10	5	0	0	0	0
48	145	18	10	0	7	0	0
49	99	16	8	0	2	0	0
50	68	19	10	0	0	0	0
51	104	17	9	0	3	0	0
52	96	17	10	0	3	0	0
53	116	22	12	0	2	0	0
54	76	18	9	0	0	0	0
55	76	14	7	0	0	0	0
56	78	19	10	0	0	0	0
57	69	19	10	0	0	0	0
58	104	22	11	0	0	0	0
59	105	20	11	0	5	0	0
60	56	11	6	0	0	0	0
61	64	9	5	0	0	0	0
62	72	15	8	0	3	0	0
63	69	0	0	0	1	0	0
64	88	13	8	0	1	0	0
65	69	13	7	0	0	0	0
66	127	21	12	0	5	0	0
67	101	6	7	0	1	0	0
68	86	11	8	0	1	0	0
69	78	5	3	0	0	0	0
70	102	13	8	0	4	0	0
71	86	4	3	0	1	0	0
72	68	9	5	0	0	0	0
73	88	7	4	0	0	0	0
74	103	12	7	0	2	0	0
75	108	10	8	0	3	0	0

August 19-21 2007– London:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area

						(H-B)	
1	75	16	8	0	0	0	0
2	69	14	7	0	0	0	0
3	82	18	9	0	0	0	0
4	101	23	12	0	3	0	0
5	84	7	4	0	1	0	0
6	108	15	9	0	5	0	0
7	89	0	0	0	1	0	0
8	77	18	9	0	0	0	0
9	87	20	9	0	0	0	0
10	86	4	4	0	0	0	0
11	107	19	9	0	2	0	0
12	95	12	6	0	5	0	0
13	75	20	10	0	0	0	0
14	86	20	11	0	0	0	0
15	81	22	12	0	0	0	0
16	77	20	10	0	0	0	0
17	83	13	7	0	0	0	0
18	95	14	8	0	0	0	0
19	90	17	9	0	1	0	0
20	123	18	11	0	3	0	0
21	118	21	9	0	4	0	0
22	74	17	9	0	2	0	0
23	108	22	11	0	0	0	0
24	79	18	9	0	0	0	0
25	115	21	11	0	1	0	0
26	99	2	1	0	7	0	0
27	132	27	14	0	2	0	0
28	97	18	10	0	1	0	0
29	112	24	11	0	2	0	0
30	114	20	9	0	1	0	0
31	97	19	10	0	2	0	0
32	99	20	10	0	6	0	0
33	114	21	11	0	1	0	0
34	100	7	5	0	0	0	0
35	91	13	8	0	2	0	0
36	115	22	12	0	0	0	0
37	109	26	13	0	5	0	0
38	129	25	13	0	2	0	0
39	112	22	11	0	0	0	0
40	100	20	11	0	0	0	0
41	115	27	14	0	0	0	0
42	96	20	10	0	0	0	0
43	99	27	14	0	9	0	0
44	114	9	6	0	1	0	0
45	110	26	13	0	2	0	0
46	111	22	11	0	0	0	0
47	87	20	10	0	0	0	0

48	98	24	12	0	0	0	0
49	90	13	7	0	0	0	0
50	95	19	10	0	0	0	0
51	139	28	14	0	4	0	0
52	98	20	11	0	0	0	0
53	89	13	8	0	0	0	0
54	99	22	11	0	0	0	0
55	123	26	15	0	0	0	0
56	89	16	8	0	0	0	0
57	92	17	9	0	0	0	0
58	80	17	9	0	0	0	0
59	137	26	13	0	3	0	0
60	98	20	10	0	0	0	0
61	99	21	12	0	0	0	0
62	85	16	8	0	0	0	0
63	117	28	14	0	0	0	0
64	90	22	11	0	0	0	0
65	114	20	11	0	0	0	0
66	113	10	5	0	0	0	0
67	123	28	14	0	0	0	0
68	110	20	10	0	0	0	0
69	100	24	12	0	0	0	0
70	105	20	11	0	0	0	0
71	87	20	10	0	0	0	0
72	89	19	10	0	0	0	0
73	87	21	11	0	0	0	0
74	65	18	9	0	0	0	0
75	88	16	9	0	0	0	0

August 19-21 2007– Grand Bend:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	105	15	7	0	0	0	0
2	95	15	8	0	0	0	0
3	82	15	8	0	0	0	0
4	81	22	11	0	0	0	0
5	83	18	9	0	1	0	0
6	78	14	7	0	0	0	0
7	100	22	11	0	4	0	0
8	82	122	12	0	2	0	0
9	94	26	14	0	1	0	0
10	91	26	13	0	1	0	0
11	95	17	9	0	0	0	0
12	66	18	10	0	0	0	0
13	77	14	6	0	1	0	0
14	92	25	13	0	0	0	0
15	72	9	5	0	0	0	0

16	95	8	6	0	0	0	0
17	124	27	14	0	6	0	0
18	100	25	12	0	7	0	0
19	69	18	6	0	0	0	0
20	100	18	9	0	3	0	0
21	95	15	9	0	1	0	0
22	98	11	5	0	2	0	0
23	104	20	12	0	5	0	0
24	96	25	13	0	11	0	0
25	96	25	12	0	0	0	0
26	86	28	14	0	0	0	0
27	74	26	12	0	0	0	0
28	66	15	8	0	0	0	0
29	60	12	6	0	0	0	0
30	70	9	5	0	0	0	0
31	84	19	10	0	0	0	0
32	89	19	10	0	0	0	0
33	120	20	10	0	0	0	0
34	87	22	11	0	0	0	0
35	64	14	7	0	0	0	0
36	86	14	7	0	0	0	0
37	88	12	6	0	0	0	0
38	74	10	5	0	0	0	0
39	79	12	7	0	0	0	0
40	99	24	12	0	0	0	0
41	70	20	10	0	0	0	0
42	86	13	7	0	0	0	0
43	78	14	7	0	2	0	0
44	77	16	8	0	0	0	0
45	94	20	10	0	0	0	0
46	130	26	13	0	3	0	0
47	133	22	11	0	3	0	0
48	98	15	7	0	3	0	0
49	115	22	11	0	2	0	0
50	94	19	10	0	0	0	0
51	93	16	8	0	2	0	0
52	89	16	8	0	1	0	0
53	100	22	11	0	2	0	0
54	110	22	14	0	0	0	0
55	104	22	11	0	0	0	0
56	95	18	10	0	3	0	0
57	72	22	11	0	0	0	0
58	80	15	8	0	1	0	0
59	81	14	7	0	0	0	0
60	88	15	8	0	0	0	0
61	79	14	7	0	0	0	0
62	124	18	13	0	3	0	0
63	85	21	11	0	0	0	0

64	89	22	11	0	0	0	0
65	100	17	9	0	2	0	0
66	98	14	8	0	3	0	0
67	105	16	8	0	2	0	0
68	94	24	12	0	0	0	0
69	88	22	11	0	0	0	0
70	89	18	9	0	0	0	0
71	100	22	11	0	0	0	0
72	79	16	8	0	0	0	0
73	79	13	7	0	0	0	0
74	111	24	12	0	2	0	0
75	110	24	12	0	4	0	0

August 19-21 2007– Sarnia:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	97	18	9	0	4	0	0
2	100	17	9	0	4	0	0
3	87	24	12	0	2	0	0
4	128	24	13	0	10	0	0
5	126	28	15	0	6	0	0
6	113	30	15	0	3	0	0
7	103	23	12	0	5	0	0
8	92	18	9	0	2	0	0
9	95	26	13	0	6	0	0
10	78	24	12	0	2	0	0
11	74	25	13	0	1	0	0
12	89	22	12	0	3	0	0
13	60	22	11	0	0	0	0
14	67	17	9	0	0	0	0
15	83	23	12	0	0	0	0
16	71	17	9	0	0	0	0
17	91	28	14	0	0	0	0
18	85	21	11	0	0	0	0
19	89	20	10	0	1	0	0
20	92	23	12	0	2	0	0
21	73	18	9	0	0	0	0
22	77	24	12	0	1	0	0
23	100	20	10	0	3	0	0
24	86	18	9	0	4	0	0
25	96	26	13	0	2	0	0
26	107	24	12	0	6	0	0
27	101	23	12	0	5	0	0
28	65	18	9	0	0	0	0
29	56	16	8	0	0	0	0
30	96	28	14	0	0	0	0

31	106	28	14	0	4	0	0
32	59	16	8	0	0	0	0
33	53	16	8	0	0	0	0
34	73	18	9	0	0	0	0
35	93	31	17	0	0	0	0
36	123	32	17	0	1	0	0
37	78	17	9	0	0	0	0
38	80	18	9	0	0	0	0
39	77	15	8	0	0	0	0
40	75	18	9	0	0	0	0
41	105	24	12	0	4	0	0
42	100	22	11	0	3	0	0
43	76	17	9	0	0	0	0
44	91	19	10	0	0	0	0
45	80	23	12	0	0	0	0
46	79	15	8	0	0	0	0
47	99	17	9	0	0	0	0
48	89	16	8	0	0	0	0
49	82	17	9	0	0	0	0
50	76	25	13	0	0	0	0
51	77	23	12	0	0	0	0
52	88	19	10	0	0	0	0
53	87	18	9	0	0	0	0
54	90	24	12	0	0	0	0
55	72	21	11	0	0	0	0
56	97	19	11	0	4	0	0
57	86	20	10	0	0	0	0
58	116	17	9	0	7	0	0
59	115	18	9	0	8	0	0
60	111	19	10	0	0	0	0
61	88	18	9	0	0	0	0
62	89	20	10	0	0	0	0
63	110	11	7	0	4	0	0
64	100	26	13	0	0	0	0
65	100	9	7	0	3	0	0
66	115	18	9	0	0	0	0
67	73	15	8	0	0	0	0
68	88	18	9	0	0	0	0
69	80	14	7	0	0	0	0
70	82	18	9	0	0	0	0
71	110	22	11	0	0	0	0
72	81	18	10	0	0	0	0
73	100	17	9	0	3	0	0
74	87	14	7	0	0	0	0
75	89	14	7	0	0	0	0

August 19-21 2007– Essex:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	100	23	12	0	3	0	0
2	73	18	9	0	0	10	1
3	130	24	12	0	3	0	0
4	94	21	11	0	3	0	0
5	97	15	10	0	6	7	1
6	90	17	11	0	3	4	1
7	83	20	10	0	2	4	1
8	87	22	13	0	2	10	1
9	76	20	10	0	0	6	1
10	87	26	13	0	0	0	0
11	86	23	12	0	0	0	0
12	89	17	9	0	2	0	0
13	98	23	12	0	0	0	0
14	96	24	12	0	1	0	0
15	86	26	13	0	0	0	0
16	86	24	13	0	0	0	0
17	83	27	14	0	0	0	0
18	95	21	11	0	2	0	0
19	82	23	12	0	0	0	0
20	90	21	11	0	3	0	0
21	77	24	12	0	0	0	0
22	105	24	13	0	4	0	0
23	94	19	10	0	2	0	0
24	88	20	11	0	1	0	0
25	86	17	10	0	0	0	0
26	106	18	9	0	6	0	0
27	96	23	12	0	2	0	0
28	94	16	8	0	1	0	0
29	105	22	12	0	2	0	0
30	123	22	11	0	2	7	1
31	117	18	9	0	6	0	0
32	124	16	9	0	4	1	1
33	121	17	9	0	2	0	0
34	123	21	11	0	6	0	0
35	106	19	11	0	4	0	0
36	100	22	12	0	3	1	1
37	86	19	11	0	1	0	0
38	119	25	14	0	4	0	0
39	116	18	9	0	2	0	0
40	117	29	13	0	4	0	0
41	117	19	11	0	2	3	1
42	111	19	10	0	2	0	0
43	86	11	6	0	2	1	1
44	100	22	12	0	1	0	0
45	94	20	12	0	2	3	1
46	100	22	12	0	2	0	0
47	109	18	9	0	4	0	0

48	84	20	11	0	1	1	1
49	85	20	10	0	0	2	1
50	100	33	17	0	3	0	0
51	122	20	11	0	6	0	0
52	123	22	12	0	4	0	0
53	98	17	9	0	3	0	0
54	83	20	10	0	2	0	0
55	90	21	11	0	0	4	1
56	110	19	11	0	1	0	0
57	98	15	8	0	1	0	0
58	79	19	10	0	0	0	0
59	100	23	13	0	5	2	1
60	132	24	13	0	8	0	0
61	82	18	10	0	2	0	0
62	99	16	8	0	2	0	0
63	99	18	8	0	1	0	0
64	96	19	11	0	4	1	1
65	94	14	7	0	1	0	0
66	113	21	11	0	2	0	0
67	112	21	11	0	3	0	0
68	120	23	13	0	6	0	0
69	84	20	10	0	0	0	0
70	83	22	11	0	0	0	0
71	84	17	9	0	0	0	0
72	90	19	10	0	1	0	0
73	90	28	15	0	1	5	1
74	87	20	10	0	0	0	0
75	104	21	11	0	0	5	1

August 19-21 2007– Chatham:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	66	12	6	0	0	0	0
2	77	12	6	0	1	0	0
3	110	14	8	0	3	0	0
4	90	17	9	0	1	0	0
5	96	16	8	0	0	0	0
6	71	14	8	0	0	0	0
7	82	17	9	0	0	0	0
8	66	19	10	0	0	0	0
9	56	12	6	0	0	0	0
10	65	15	8	0	0	0	0
11	65	10	5	0	0	0	0
12	57	13	7	0	0	0	0
13	70	17	9	0	0	0	0
14	91	18	10	0	0	0	0
15	90	16	8	0	0	0	0
16	56	12	6	0	0	0	0
17	61	9	5	0	0	0	0
18	56	12	7	0	0	0	0
19	52	10	5	0	0	0	0

20	61	9	7	0	0	0	0
21	63	14	7	0	0	0	0
22	63	20	11	0	0	0	0
23	78	20	10	0	0	0	0
24	71	12	6	0	0	0	0
25	82	11	6	0	0	0	0
26	66	26	13	0	0	0	0
27	58	18	10	0	0	0	0
28	54	7	4	0	0	0	0
29	73	19	10	0	0	0	0
30	113	22	11	0	1	0	0
31	125	18	10	0	3	0	0
32	124	14	7	0	2	0	0
33	113	19	10	0	3	0	0
34	86	14	7	0	0	2	0
35	88	16	8	0	0	0	0
36	66	14	7	0	0	0	0
37	90	21	11	0	0	2	1
38	61	18	9	0	0	0	0
39	89	16	8	0	0	0	0
40	98	20	10	0	0	0	0
41	71	24	12	0	0	0	0
42	97	20	13	0	0	0	0
43	69	11	6	0	0	0	0
44	99	28	14	0	0	0	0
45	100	24	12	0	0	0	0
46	95	25	13	0	0	0	0
47	90	24	12	0	0	0	0
48	96	28	13	0	0	0	0
49	73	19	12	0	0	0	0
50	75	24	12	0	0	0	0
51	100	27	14	0	2	0	0
52	100	30	15	0	0	0	0
53	110	30	16	0	0	0	0
54	100	28	14	0	0	0	0
55	92	26	13	0	0	0	0
56	74	16	8	0	0	0	0
57	56	13	7	0	0	0	0
58	66	17	9	0	0	0	0
59	83	26	13	0	0	0	0
60	100	20	11	0	0	0	0
61	83	17	9	0	0	0	0
62	86	19	10	0	0	0	0
63	86	11	6	0	0	0	0
64	89	14	7	0	0	0	0
65	75	12	6	0	0	0	0
66	80	14	7	0	0	0	0
67	77	17	9	0	0	0	0
68	66	17	9	0	0	0	0
69	107	25	13	0	0	6	1
70	76	16	8	0	0	6	1

71	84	22	11	0	0	0	0
72	97	22	11	0	0	0	0
73	97	18	10	0	0	0	0
74	100	25	13	0	0	0	0
75	97	17	9	0	0	0	0

August 19-21 2007– Port Stanley:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	96	17	9	0	0	0	0
2	115	14	7	0	0	0	0
3	100	16	8	0	0	0	0
4	114	13	8	0	2	0	0
5	98	11	6	0	0	0	0
6	100	11	6	0	0	0	0
7	107	17	9	0	1	0	0
8	110	13	7	0	0	0	0
9	110	16	8	0	1	0	0
10	104	12	6	0	0	0	0
11	110	16	8	0	3	0	0
12	98	14	7	0	1	0	0
13	112	12	7	0	3	0	0
14	110	11	6	0	2	0	0
15	110	12	6	0	1	0	0
16	113	10	6	0	1	0	0
17	99	11	6	0	1	0	0
18	98	15	8	0	1	0	0
19	10	9	5	0	0	0	0
20	99	11	6	0	0	0	0
21	95	10	5	0	0	0	0
22	99	11	6	0	0	0	0
23	91	10	6	0	1	0	0
24	94	14	8	0	0	0	0
25	96	14	8	0	0	0	0
26	102	12	7	0	1	0	0
27	101	21	11	0	3	0	0
28	112	16	8	0	2	0	0
29	105	17	9	0	0	0	0
30	115	17	9	0	0	0	0
31	122	17	10	0	4	0	0
32	123	22	11	0	0	0	0
33	105	17	10	0	1	0	0
34	110	20	11	0	2	0	0
35	110	20	10	0	0	0	0
36	120	19	11	0	0	0	0
37	112	11	8	0	0	0	0
38	122	17	9	0	0	0	0
39	111	19	10	0	0	0	0
40	135	16	10	0	5	0	0
41	126	22	12	0	2	0	0
42	163	23	13	0	13	0	0

43	130	22	11	0	0	0	0
44	111	18	9	0	0	0	0
45	94	18	9	0	0	0	0
46	123	23	12	0	0	0	0
47	137	22	11	0	0	0	0
48	139	20	10	0	0	0	0
49	140	12	8	0	5	0	0
50	158	16	8	0	4	0	0
51	146	26	13	0	0	0	0
52	138	19	10	0	0	0	0
53	140	22	11	0	0	0	0
54	98	18	9	0	0	0	0
55	116	22	11	0	0	0	0
56	119	24	12	0	0	0	0
57	146	22	12	0	1	0	0
58	150	22	11	0	2	0	0
59	98	16	8	0	0	0	0
60	92	15	9	0	0	0	0
61	127	19	11	0	0	0	0
62	115	20	10	0	0	0	0
63	96	14	8	0	0	0	0
64	101	14	7	0	0	0	0
65	81	8	5	0	0	0	0
66	137	24	13	0	2	0	0
67	100	14	7	0	0	0	0
68	152	12	6	0	0	0	0
69	108	16	8	0	0	0	0
70	124	21	11	0	0	0	0
71	140	22	11	0	3	0	0
72	117	20	11	0	0	0	0
73	122	19	10	0	0	0	0
74	126	17	9	0	0	0	0
75	140	24	12	0	0	0	0

August 19-21 2007– Simcoe:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	92	18	9	0	5	0	0
2	96	26	13	0	0	0	0
3	108	18	10	0	4	0	0
4	100	16	8	0	3	0	0
5	132	20	10	0	7	0	0
6	136	18	9	0	11	0	0
7	134	22	11	0	5	0	0
8	140	23	12	0	11	0	0
9	114	16	9	0	6	0	0
10	137	23	12	0	10	0	0
11	86	17	9	0	0	0	0
12	132	22	11	0	6	0	0
13	131	24	12	0	9	0	0
14	130	19	10	0	12	0	0

15	126	17	9	0	8	0	0
16	98	23	12	0	2	0	0
17	99	17	9	0	6	0	0
18	99	16	8	0	4	0	0
19	122	19	11	0	11	0	0
20	121	21	11	0	8	0	0
21	100	17	9	0	11	0	0
22	70	16	8	0	0	0	0
23	106	20	10	0	7	0	0
24	96	20	11	0	5	0	0
25	98	21	11	0	9	0	0
26	78	14	7	0	0	0	0
27	90	16	8	0	0	0	0
28	77	16	8	0	0	0	0
29	81	16	8	0	6	0	0
30	105	20	10	0	11	0	0
31	86	14	8	0	0	0	0
32	75	20	10	0	0	0	0
33	130	24	12	0	15	0	0
34	127	22	11	0	8	0	0
35	117	23	13	0	12	0	0
36	108	14	8	0	7	0	0
37	90	19	10	0	3	0	0
38	90	15	8	0	5	0	0
39	91	20	10	0	6	0	0
40	120	23	12	0	11	0	0
41	79	17	9	0	0	0	0
42	128	22	11	0	9	0	0
43	118	10	5	0	1	0	0
44	115	26	13	0	10	0	0
45	117	16	8	0	2	0	0
46	62	18	10	0	0	0	0
47	95	17	9	0	0	0	0
48	92	21	11	0	0	0	0
49	100	26	13	0	0	0	0
50	107	17	10	0	0	0	0
51	117	22	12	0	0	0	0
52	82	16	8	0	0	0	0
53	108	20	11	0	3	0	0
54	80	14	8	0	0	0	0
55	122	22	11	0	0	0	0
56	99	19	11	0	0	0	0
57	94	20	10	0	3	0	0
58	110	19	11	0	1	0	0
59	130	20	10	0	0	0	0
60	119	24	13	0	1	0	0
61	114	22	11	0	0	0	0
62	130	29	15	0	3	0	0
63	111	26	14	0	1	0	0
64	140	22	12	0	0	0	0
65	93	16	8	0	0	0	0

66	94	17	9	0	0	0	0
67	86	21	11	0	0	0	0
68	82	20	10	0	0	0	0
69	104	22	11	0	0	0	0
70	99	19	10	0	0	0	0
71	112	17	9	0	0	0	0
72	116	19	11	0	1	0	0
73	88	21	12	0	0	0	0
74	89	17	9	0	0	0	0
75	99	21	11	0	1	0	0

August 26-28 2007– Brantford:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	77	16	8	0	0	14	2.3
2	117	17	9		3	0	0
3	70	9	5		0	0	0
4	89	7	4		0	0	0
5	96	11	6		3	0	0
6	77	6	4		2	0	0
7	67	3	3		0	0	0
8	75	4	3		0	0	0
9	95	18	9		3	0	0
10	115	14	7		6	0	0
11	122	10	6		4	0	0
12	111	12	6		3	0	0
13	120	11	7		3	0	0
14	112	10	6		4	0	0
15	113	0	2		0	0	0
16	128	3	3		3	0	0
17	137	14	7		7	0	0
18	67	10	5		0	0	0
19	96	18	9		0	0	0
20	67	10	6		0	0	0
21	68	5	4		0	0	0
22	83	16	8		0	0	0
23	74	9	6		0	0	0
24	102	24	12		0	0	0
25	100	19	10		0	0	0
26	100	18	9		0	0	0
27	87	12	6		0	0	0
28	69	10	6		0	0	0
29	95	5	5		0	0	0
30	110	17	9		0	0	0
31	95	7	5		2	0	0
32	84	17	9		0	0	0
33	47	18	9		0	0	0
34	86	17	10		0	8	1.6
35	76	17	9		0	1	1
36	55	14	7		0	0	0
37	88	12	6		0	0	0

38	116	17	9		0	0	0
39	87	10	7		1	0	0
40	78	17	9		0	0	0
41	99	10	7		0	0	0
42	90	13	7		1	0	0
43	94	15	8		3	0	0
44	74	15	8		1	0	0
45	86	21	11		0	0	0
46	60	11	6		0	0	0
47	80	14	7		0	0	0
48	76	12	6		0	0	0
49	70	10	5		0	0	0
50	125	25	13		4	0	0
51	61	18	9		0	0	0
52	98	9	5		0	0	0
53	120	9	5		0	0	0
54	72	18	9		0	0	0
55	60	17	9		0	0	0
56	100	14	7		1	0	0
57	120	17	9		3	0	0
58	99	17	9		3	0	0
59	114	23	12		2	0	0
60	81	19	10		0	0	0
61	78	18	9		0	0	0
62	77	14	7		0	0	0
63	72	17	9		0	0	0
64	107	20	11		5	0	0
65	77	11	6		0	0	0
66	63	8	5		0	0	0
67	70	15	9		3	0	0
68	87	13	7		1	0	0
69	126	21	11		5	1	1
70	99	7	7		1	0	0
71	89	12	7		1	0	0
72	105	12	7		2	0	0
73	100	12	7		0	0	0
74	105	8	5		0	0	0
75	103	14	8		3	0	0

August 26-28 2007– London:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	100	24	13	0	3	0	0
2	66	14	7		0	5	1
3	73	16	8		0	0	0
4	78	16	9		0	0	0
5	102	14	8		0	0	0
6	85	7	4		1	0	0
7	71	15	8		0	0	0
8	85	19	9		0	0	0
9	80	6	4		0	0	0

10	107	19	9	2	0	0
11	98	12	6	5	0	0
12	81	26	13	0	0	0
13	66	14	8	0	0	0
14	76	19	10	0	0	0
15	93	20	11	0	0	0
16	84	21	11	0	0	0
17	80	22	11	0	0	0
18	81	13	7	0	0	0
19	92	14	8	0	0	0
20	89	14	8	1	0	0
21	95	17	9	0	0	0
22	69	17	9	0	0	0
23	122	17	17	4	0	0
24	119	20	20	4	0	0
25	100	12	12	1	0	0
26	129	12	12	9	0	0
27	108	18	18	1	0	0
28	96	18	18	2	0	0
29	120	24	24	0	0	0
30	80	17	17	1	0	0
31	100	2	2	1	0	0
32	78	9	9	0	0	0
33	110	22	22	0	1	3
34	77	16	8	0	0	0
35	120	22	11	3	2	1
36	110	23	12	2	1	1
37	130	27	14	8	6	1
38	109	25	13	1	0	0
39	113	9	6	8	1	1
40	112	22	11	2	2	1
41	111	23	12	2	2	1
42	110	26	13	0	0	0
43	110	20	10	2	0	0
44	129	26	14	5	0	0
45	114	22	11	2	0	0
46	100	21	11	0	0	0
47	120	24	15	0	0	0
48	98	26	13	0	0	0
49	116	25	13	0	0	0
50	98	22	11	0	0	0
51	97	22	11	0	0	0
52	99	27	14	0	0	0
53	88	13	8	0	0	0
54	96	24	12	0	0	0
55	90	20	10	0	0	0
56	96	20	11	0	0	0
57	99	19	12	0	0	0
58	139	30	15	4	0	0
59	86	12	7	0	0	0
60	90	15	8	0	0	0

61	91	17	9		0	0	0
62	77	19	10		0	0	0
63	134	26	13		3	0	0
64	100	19	10		0	0	0
65	97	19	12		0	0	0
66	98	22	12		0	0	0
67	90	15	8		0	0	0
68	88	19	10		0	0	0
69	108	20	11		0	0	0
70	113	16	8		0	0	0
71	120	28	14		0	0	0
72	99	17	9		0	0	0
73	101	24	12		0	0	0
74	100	21	11		0	0	0
75	81	19	10		0	0	0

August 26-28 2007– Grand Bend:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	124	26	13	0	2	0	0
2	95	16	8		3	0	0
3	127	20	10		4	0	0
4	125	24	12		4	0	0
5	82	18	10		1	0	0
6	84	16	7		2	0	0
7	66	18	9		0	0	0
8	79	18	9		0	0	0
9	66	18	9		0	0	0
10	85	22	11		0	1	1
11	80	12	6		0	0	0
12	74	12	6		1	0	0
13	76	16	8		0	0	0
14	105	24	12		0	5	1
15	94	22	11		0	0	0
16	105	26	13		2	1	1
17	90	25	13		0	0	0
18	78	18	9		0	1	1
19	120	20	5		0	8	1
20	83	18	9		0	0	0
21	84	18	9		0	0	0
22	64	9	5		0	0	0
23	52	12	6		0	0	0
24	63	14	8		0	0	0
25	82	20	10		0	0	0
26	69	22	11		0	0	0
27	91	17	10		3	0	0
28	100	23	13		2	0	0
29	100	22	11		0	2	1
30	79	23	12		0	0	0
31	99	12	8		1	8	1.4
32	79	14	7		1	0	0

33	78	14	7		0	0	0
34	121	18	11		3	4	1
35	125	16	11		2	4	3
36	95	17	9		2	0	1
37	87	18	10		3	0	1
38	91	24	13		4	0	0
39	74	19	12		0	4	1
40	90	20	12		1	4	1
41	120	28	14		3	1	1.3
42	100	21	11		0	0	1
43	79	8	7		0	0	0
44	119	20	11		1	0	0
45	120	12	8		2	0	0
46	139	17	11		0	0	0
47	155	19	10		7	0	0
48	126	17	9		1	0	0
49	64	20	10		0	0	0
50	118	15	8		2	0	0
51	120	23	12		1	0	0
52	86	23	12		0	0	0
53	68	18	9		0	0	0
54	118	14	8		0	0	0
55	121	20	10		2	0	0
56	128	21	11		0	0	0
57	136	9	5		7	0	0
58	138	17	9		3	0	0
59	100	16	7		0	0	0
60	116	20	10		0	0	0
61	76	20	10		0	0	0
62	132	21	11		4	0	0
63	133	16	8		2	0	0
64	123	21	11		6	0	0
65	119	24	10		2	0	0
66	124	19	10		3	0	0
67	121	16	8		0	0	0
68	73	14	7		0	0	0
69	140	20	10		2	0	0
70	123	17	9		0	0	0
71	95	20	10		0	0	0
72	94	20	10		0	0	0
73	123	18	9		2	0	0
74	128	21	11		5	0	0
75	111	13	7		0	0	0

August 26-28 2007– Sarnia:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	94	18	10	0	4	0	0
2	100	19	10		4	0	0
3	92	17	9		2	0	0
4	100	23	12		5	0	0

5	125	25	13	11	0	0
6	124	28	15	7	0	0
7	109	28	14	3	0	0
8	83	20	11	1	0	0
9	104	24	12	7	0	0
10	97	26	13	6	0	0
11	74	24	12	2	0	0
12	76	25	13	1	0	0
13	100	26	14	3	0	0
14	65	22	11	1	0	0
15	74	23	12	0	0	0
16	87	25	12	0	0	0
17	99	28	14	0	0	0
18	71	17	9	0	0	0
19	92	20	10	1	0	0
20	85	23	12	0	0	0
21	91	22	12	2	0	0
22	75	20	10	0	0	0
23	89	23	12	0	0	0
24	76	21	11	0	0	0
25	86	18	10	0	0	0
26	97	29	15	0	0	0
27	132	34	18	0	0	0
28	101	33	18	0	0	0
29	79	20	11	0	0	0
30	60	16	8	0	0	0
31	112	30	15	4	0	0
32	100	20	10	5	0	0
33	98	26	13	8	0	0
34	64	17	9	0	0	0
35	75	22	11	1	0	0
36	87	17	9	4	0	0
37	104	22	11	6	0	0
38	98	25	13	4	0	0
39	90	18	9	0	1	1
40	82	16	8	0	0	0
41	89	17	9	0	0	0
42	82	17	9	0	0	0
43	68	19	10	0	0	0
44	95	23	12	0	0	0
45	95	21	11	3	0	0
46	106	24	12	4	4	1.6
47	98	22	11	2	2	3
48	95	17	10	0	0	0
49	74	17	9	0	0	0
50	78	25	13	0	0	0
51	71	21	11	0	0	0
52	95	18	9	1	0	0
53	81	18	9	0	0	0
54	71	24	12	0	0	0
55	85	20	10	0	0	0

56	111	16	8		8	0	0
57	115	17	9		5	0	0
58	87	19	10		0	0	0
59	87	20	10		0	0	0
60	73	14	7		0	0	0
61	79	18	9		0	0	0
62	109	15	8		9	0	0
63	102	20	10		3	0	0
64	100	11	7		4	0	0
65	99	26	13		0	0	0
66	69	14	7		0	0	0
67	99	9	7		3	0	0
68	107	16	9		2	0	0
69	90	14	7		1	0	0
70	99	13	9		0	0	0
71	100	7	5		4	0	0
72	117	17	9		5	0	0
73	82	18	9		0	0	0
74	68	17	9		0	0	0
75	74	16	8		0	0	0

August 26-28 2007– Essex:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	71	21	11	0	1	0	0
2	60	21	11		0	0	0
3	65	26	13		1	0	0
4	61	16	8		0	0	0
5	69	20	11		0	0	0
6	76	20	10		0	0	0
7	77	17	9		0	0	0
8	98	26	13		0	0	0
9	90	23	13		2	15	1.3
10	100	13	9		6	7	1.8
11	86	23	12		2	0	0
12	93	21	11		0	0	0
13	120	24	14		7	17	1
14	76	20	11		0	0	0
15	83	26	13		0	0	0
16	93	21	11		1	0	0
17	90	28	14		0	1	1
18	97	21	11		2	5	1
19	98	12	6		3	0	0
20	88	18	11		0	0	0
21	104	21	12		2	11	1
22	121	22	11		6	4	1
23	87	19	10		7	0	0
24	123	17	9		7	2	2.5
25	74	24	12		1	0	0
26	100	22	11		5	7	1
27	100	18	10		4	1	1

28	111	24	11	6	0	1
29	120	20	11	8	0	0
30	94	15	10	1	0	0
31	89	15	8	3	0	0
32	123	19	10	8	3	1
33	93	17	9	2	0	0
34	94	12	6	5	6	1
35	93	17	9	2	0	0
36	82	13	7	3	0	0
37	110	27	14	6	0	0
38	102	15	9	3	0	0
39	100	16	1	9	0	0
40	96	17	12	4	0	0
41	94	25	13	7	2	1
42	88	21	11	10	9	1
43	86	24	12	4	5	1
44	72	11	8	2	9	1.42
45	61	14	7	0	8	2.75
46	99	18	9	2	12	1.75
47	107	9	7	6	9	1
48	93	12	6	3	8	1.6
49	85	16	8	2	8	1.1
50	86	10	6	0	4	1.5
51	74	22	11	0	0	0
52	99	20	10	2	0	0
53	108	18	10	2	2	1
54	77	20	10	0	0	0
55	97	23	12	2	0	0
56	100	17	9	4	2	1
57	75	22	11	0	13	1
58	76	22	11	0	12	1
59	84	18	9	2	0	0
60	106	21	11	5	13	1.3
61	82	19	10	0	0	0
62	112	22	11	6	0	0
63	98	21	11	0	0	0
64	80	21	11	0	0	0
65	106	20	10	3	0	0
66	99	18	9	3	0	0
67	96	20	11	5	0	0
68	87	19	10	4	0	0
69	100	26	14	5	0	0
70	91	18	10	3	0	0
71	90	22	11	1	0	0
72	99	22	12	5	0	0
73	100	22	11	4	0	0
74	91	15	8	2	0	0
75	111	20	13	2	0	0

August 26-28 2007– Chatham:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area

						(H-B)	
1	80	13	6	0	1	0	0
2	65	11	6		0	0	0
3	109	14	8		3	0	0
4	88	17	9		1	0	0
5	96	16	8		0	0	0
6	71	15	8		0	0	0
7	82	15	8		0	0	0
8	64	15	8		0	0	0
9	64	15	8		0	0	0
10	64	14	7		0	0	0
11	65	9	5		0	0	0
12	95	8	7		0	8	1
13	88	18	9		0	0	0
14	57	15	8		0	0	0
15	72	15	8		0	0	0
16	55	10	7		0	0	0
17	52	10	6		0	0	0
18	53	11	6		0	0	0
19	90	27	15		0	14	1.7
20	67	25	13		0	6	2
21	68	18	9		0	0	0
22	78	25	13		0	2	1.5
23	79	7	5		0	0	0
24	61	26	14		0	15	1.5
25	60	12	6		0	10	3.4
26	58	15	8		0	0	0
27	53	8	5		0	5	2.3
28	76	18	10		0	0	0
29	105	21	11		1	0	0
30	121	17	10		3	0	0
31	116	10	6		2	0	0
32	84	16	8		0	0	0
33	78	17	10		0	5	3
34	83	27	14		0	10	2.3
35	52	18	9		0	8	1
36	55	18	9		0	4	1.3
37	83	14	9		0	9	2.1
38	100	22	12		0	0	0
39	63	23	12		0	0	0
40	92	19	13		0	0	0
41	72	11	6		0	0	0
42	95	28	14		0	0	0
43	98	26	13		0	0	0
44	94	24	13		0	0	0
45	90	24	13		0	0	0
46	97	28	14		0	0	0
47	67	21	12		0	0	0
48	73	25	13		0	0	0
49	72	19	10		0	0	0
50	105	31	16		0	0	0

51	112	22	12		0	0	0
52	106	28	15		0	0	0
53	76	24	12		0	0	0
54	75	16	8		0	0	0
55	88	13	7		0	0	0
56	91	21	11		0	0	0
57	88	14	8		0	0	0
58	82	14	8		0	0	0
59	90	14	7		0	0	0
60	81	14	7		0	0	0
61	79	16	8		0	0	0
62	64	17	9		0	0	0
63	68	23	12		0	4	2
64	100	24	12		0	24	4
65	80	16	8		0	16	3.3
66	83	21	11		0	14	1.6
67	100	25	13		0	16	2.9
68	98	22	11		0	7	2.6
69	98	21	11		0	5	1.4
70	93	15	10		0	3	1.6
71	68	21	11		0	14	2
72	117	24	13		5	0	0
73	61	21	11		0	0	0
74	58	19	11		0	0	0
75	132	15	8		0	0	0

August 26-28 2007– Port Stanley:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	93	19	10	0	0	0	0
2	89	8	5		0	0	0
3	109	15	8		0	0	0
4	95	15	8		0	0	0
5	94	13	7		0	0	0
6	94	13	7		0	1	1
7	109	14	8		0	1	1
8	99	9	5		0	0	0
9	97	10	6		0	0	0
10	108	12	7		1	0	0
11	100	14	8		1	0	0
12	110	10	6		0	0	0
13	115	16	8		1	0	0
14	108	11	6		1	0	0
15	115	15	8		0	0	0
16	109	11	6		3	0	0
17	111	11	7		2	1	1
18	96	12	7		3	0	0
19	87	10	6		0	0	0
20	94	12	7		1	0	0

21	86	16	8	0	3	1
22	99	11	6	1	0	0
23	84	8	5	0	0	0
24	100	10	6	0	0	0
25	87	12	9	1	0	0
26	100	19	10	0	1	1
27	108	15	8	3	3	1
28	85	12	7	2	11	2.5
29	100	14	7	0	5	1
30	128	20	10	0	9	1.25
31	98	14	8	1	2	1
32	97	21	11	0	0	0
33	132	28	14	0	0	0
34	115	14	11	0	11	2.5
35	115	11	7	0	7	1.8
36	114	13	9	0	0	0
37	122	16	8	0	0	0
38	70	10	6	0	0	0
39	107	18	9	0	0	0
40	125	0	0	5	0	0
41	129	20	11	1	0	0
42	116	10	6	0	0	0
43	107	20	10	0	0	0
44	94	17	9	0	0	0
45	120	22	12	0	0	0
46	130	21	11	0	0	0
47	138	19	10	0	0	0
48	135	17	9	0	0	0
49	134	20	11	0	0	0
50	134	20	11	0	0	0
51	150	26	13	0	0	0
52	120	17	9	0	0	0
53	145	21	11	1	0	0
54	149	22	11	1	0	0
55	99	16	8	0	0	0
56	89	13	7	0	0	0
57	121	17	9	2	0	0
58	146	28	15	0	0	0
59	128	17	9	0	0	0
60	93	11	8	0	0	0
61	87	9	5	0	0	0
62	76	7	5	0	0	0
63	130	15	9	0	0	0
64	109	9	5	0	0	0
65	100	17	9	0	8	1
66	104	12	6	0	11	1.4
67	124	15	8	0	0	0
68	144	17	9	0	0	0

69	133	13	7		0	0	0
70	133	16	8		0	0	0
71	107	12	6		0	0	0
72	147	19	10		0	0	0
73	126	8	6		0	0	0
74	122	13	7		0	0	0
75	160	11	6		0	0	0

August 26-28 2007– Simcoe:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	67	10	5	0	0	0	0
2	77	10	5		0	4	2
3	69	17	9		0	13	1
4	72	21	11		0	16	2.75
5	60	19	10		0	16	1.5
6	51	14	8		0	9	1.4
7	96	23	12		10	6	2
8	72	20	11		0	1	1
9	68	16	8		0	11	1
10	108	19	10		5	1	2
11	127	20	10		9	8	2
12	109	19	9		9	19	1.625
13	129	20	9		0	6	2
14	139	19	11		9	8	2
15	136	23	12		10	7	1.5
16	89	22	10		6	13	2.25
17	94	27	11		0	5	1
18	114	16	9		3	5	1.25
19	99	15	8		3	1	3
20	132	19	9		7	3	1
21	139	21	11		6	5	1.5
22	63	13	7		0	0	0
23	106	19	10		6	5	1.4
24	94	21	11		0	0	0
25	98	23	12		1	0	0
26	75	17	9		0	0	0
27	119	14	8		5	4	1.5
28	92	18	9		3	6	1.5
29	95	16	8		5	6	3
30	99	19	12		3	1	1
31	96	11	6		9	3	1
32	64	16	8		0	3	1
33	108	20	10		7	10	1.6
34	82	15	8		3	3	2
35	74	14	7		0	0	0
36	93	17	8		0	0	0
37	77	17	10		0	0	0
38	82	17	8		6	6	1.5
39	100	18	10		11	9	2

40	71	17	8		0	0	0
41	122	23	12		13	6	1
42	121	20	11		8	7	2
43	127	22	12		12	11	2
44	107	13	7		5	2	2
45	90	11	7		6	2	2
46	92	19	10		4	0	0
47	77	15	8		0	0	0
48	97	20	10		4	5	1.25
49	112	15	8		7	1	2
50	118	20	11		10	2	1.5
51	120	26	15		13	10	1.8
52	132	19	11		9	12	1.28
53	114	13	5		1	0	0
54	114	16	8		2	0	0
55	125	25	14		12	12	1.42
56	60	18	10		0	0	0
57	94	15	8		0	0	0
58	92	20	10		0	0	0
59	98	18	8		0	3	1.3
60	100	24	12		0	0	0
61	90	17	9		0	0	0
62	112	20	12		0	0	0
63	104	16	9		0	0	0
64	81	14	8		0	0	0
65	107	18	10		2	0	0
66	96	20	11		0	0	0
67	100	21	11		0	0	0
68	89	20	10		0	0	0
69	128	17	9		4	3	2
70	121	22	11		1	0	0
71	105	24	13		0	0	0
72	106	22	10		0	0	0
73	124	28	14		1	2	1
74	123	12	6		3	3	1
75	138	23	13		2	2	1.5

September 04-05 – Brantford:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	82	14	7	0	0	14	2.8
2	112	16	8		3	0	0
3	89	7	4		0	0	0
4	72	9	5		0	0	0
5	100	9	5		3	0	0
6	75	11	6		0	0	0
7	87	12	6		2	0	0
8	79	6	4		1	0	0
9	74	3	3		0	0	0
10	74	3	3		0	0	0
11	120	18	9		2	0	0

12	123	6	4	4	0	0
13	112	15	9	6	0	0
14	122	12	6	3	5	1
15	129	9	6	2	4	1
16	120	9	6	3	1	1
17	112	0	0	2	0	0
18	134	13	7	7	2	1
19	136	3	3	3	0	0
20	98	17	9	0	0	0
21	87	16	8	0	0	0
22	74	10	6	0	0	0
23	115	22	11	1	1	1
24	107	18	9	0	1	3
25	103	15	8	0	0	0
26	96	5	3	0	0	0
27	67	5	4	0	0	0
28	102	4	4	2	2	2
29	82	16	10	0	0	0
30	73	17	9	0	0	0
31	78	12	7	0	0	0
32	73	18	10	0	1	1
33	91	12	6	0	6	1.1
34	118	18	9	1	1	1
35	88	10	7	0	0	0
36	76	15	10	0	0	0
37	100	10	8	1	0	0
38	85	14	7	3	0	0
39	92	16	8	1	0	0
40	67	16	8	1	1	1
41	86	22	12	0	0	0
42	54	11	7	0	0	0
43	67	17	9	0	0	0
44	78	12	6	0	0	0
45	110	26	14	4	0	0
46	64	18	9	0	0	0
47	96	9	5	0	0	0
48	102	9	5	0	0	0
49	64	10	5	0	0	0
50	65	19	10	0	0	0
51	74	19	10	0	2	1
52	80	20	11	0	0	0
53	70	14	7	1	0	0
54	71	17	9	0	0	0
55	97	14	8	3	2	1.5
56	116	16	9	3	2	1
57	114	22	11	2	1	1
58	110	13	11	4	0	0
59	66	10	6	0	0	0
60	57	9	5	0	0	0
61	66	15	8	3	0	0
62	88	13	8	1	1	1

63	124	21	12		3	5	1
64	100	7	7		1	0	0
65	85	12	8		0	2	1.5
66	74	5	3		0	1	1
67	110	13	8		3	0	0
68	82	4	3		1	1	1
69	71	9	5		0	0	0
70	100	10	5		2	0	0
71	90	9	5		0	1	1
72	94	13	7		3	2	1.5
73	82	11	7		0	0	0
74	87	12	7		2	3	1.5
75	76	14	7		3	2	2.5

September 04-05 – London:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	100	25	12	0	3	4	1.5
2	63	14	7		0	5	1.2
3	72	16	8		0	0	0
4	75	14	7		0	0	0
5	80	7	4		1	0	0
6	81	13	7		1	1	1
7	71	16	8		0	0	0
8	84	20	9		0	0	0
9	103	13	8		6	7	1
10	80	18	9		0	0	0
11	63	14	9		0	0	0
12	78	5	4		0	0	0
13	99	16	9		3	1	2
14	97	12	6		5	3	1
15	75	21	11		0	2	1
16	127	8	5		8	1	1
17	99	13	6		1	1	1
18	119	20	9		3	4	1
19	115	16	10		4	2	1
20	89	19	10		0	0	0
21	69	11	7		0	0	0
22	79	20	10		0	0	0
23	76	18	9		1	1	1
24	85	21	11		0	0	0
25	85	20	11		0	0	0
26	81	4	3		0	0	0
27	84	13	7		0	1	1
28	76	18	9		0	0	0
29	78	17	8		0	0	0
30	87	13	8		1	4	1.75
31	92	16	9		1	0	0
32	69	17	9		0	0	0
33	87	20	10		1	0	0
34	97	19	9		2	1	1

35	86	9	5		0	0	0
36	99	2	2		1	0	0
37	112	24	11		0	0	0
38	110	18	8		1	0	0
39	98	16	8		1	1	1
40	84	18	10		0	0	0
41	100	15	10		2	1	1
42	83	11	6		0	0	0
43	111	20	10		0	1	1
44	99	7	5		1	0	0
45	78	8	6		0	0	0
46	78	16	9		0	1	1
47	69	9	5		0	0	0
48	66	21	11		0	0	0
49	86	17	9		0	0	0
50	83	15	8		0	0	0
51	71	10	7		0	0	0
52	120	12	8		7	3	1.5
53	112	21	11		2	4	1.6
54	70	10	5		0	6	1
55	129	27	14		7	6	1
56	107	21	11		2	0	0
57	78	16	9		0	0	0
58	111	25	13		1	6	1
59	114	22	11		1	10	1
60	93	20	9		0	0	0
61	93	24	12		0	0	0
62	90	13	8		0	0	0
63	100	27	14		0	2	1
64	137	28	14		5	2	2
65	100	21	11		0	0	0
66	120	27	14		0	0	0
67	124	25	14		0	0	0
68	101	20	11		0	0	0
69	108	20	11		2	0	0
70	130	24	14		5	0	0
71	99	23	12		0	0	0
72	73	12	8		0	0	0
73	97	14	8		0	0	0
74	104	21	11		0	0	0
75	112	30	15		0	0	0

September 04-05 – Grand Bend:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	93	16	8	0	0	14	1.5
2	128	23	12		3	5	1.5
3	116	18	9		0	8	1.4
4	128	23	12		3	0	0
5	113	17	9		0	11	2.67
6	79	17	9		1	13	2.4

7	82	15	8	0	9	2.6
8	77	18	9	0	4	1
9	101	24	13	0	24	1
10	100	25	13	0	2	2
11	79	20	11	0	0	0
12	68	18	10	0	7	1.2
13	83	20	10	0	5	1
14	108	22	12	0	13	1
15	118	20	10	6	8	1
16	78	19	10	0	0	0
17	82	24	14	0	0	0
18	61	13	7	0	0	0
19	66	20	10	0	0	0
20	98	17	9	3	3	1
21	74	15	8	1	6	1
22	76	14	7	0	6	1
23	87	15	8	0	9	1.8
24	76	13	7	0	13	1.5
25	122	18	11	3	6	1
26	129	18	12	2	11	1.14
27	98	13	8	1	9	1.1
28	86	0	0	1	0	0
29	100	17	10	1	6	1
30	100	22	13	0	11	0
31	89	24	12	0	0	0
32	79	21	11	0	9	2.6
33	116	28	14	3	7	1.75
34	91	18	10	2	0	0
35	100	21	13	4	10	1.1
36	72	21	12	0	13	2.3
37	89	20	12	0	14	2.7
38	113	22	12	1	0	0
39	110	15	8	2	0	0
40	66	15	8	0	0	0
41	117	22	11	0	0	0
42	84	17	9	0	0	0
43	107	17	9	2	0	0
44	123	8	5	6	0	0
45	112	18	9	0	0	0
46	97	18	8	0	0	0
47	100	21	11	1	0	0
48	115	21	11	0	0	0
49	117	15	8	1	0	0
50	70	20	10	0	0	0
51	120	23	12	0	0	0
52	87	10	5	0	0	0
53	117	19	10	0	0	0
54	113	16	8	0	0	0
55	129	18	9	0	0	0
56	117	8	4	3	0	0
57	115	18	10	0	0	0

58	97	19	10		0	0	0
59	92	20	10		0	0	0
60	100	12	7		0	0	0
61	93	14	8		0	0	0
62	100	17	9		0	0	0
63	122	18	10		2	0	0
64	122	23	11		5	0	0
65	104	15	8		0	0	0
66	108	16	8		1	0	0
67	100	15	8		0	0	0
68	120	20	10		0	0	0
69	98	13	6		0	0	0
70	129	15	8		1	0	0
71	118	17	9		0	0	0
72	100	16	8		0	0	0
73	97	15	8		0	0	0
74	96	13	7		0	0	0
75	118	12	7		0	0	0

September 04-05 – Sarnia:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	98	27	14	0	3	0	0
2	75	25	13		1	0	0
3	76	24	12		2	0	0
4	94	26	13		4	0	0
5	103	26	13		4	1	1
6	80	22	11		2	0	0
7	100	23	12		6	2	1
8	106	29	15		0	1	1
9	89	15	8		2	0	0
10	81	17	9		4	0	0
11	100	18	10		4	1	1
12	124	29	15		6	1	1
13	124	25	13		10	1	1
14	95	18	9		4	1	1
15	81	23	12		0	0	0
16	89	26	12		0	0	0
17	74	19	10		0	0	0
18	98	32	16		0	0	0
19	84	26	13		0	0	0
20	87	20	10		1	0	0
21	89	20	11		2	0	0
22	75	20	11		0	0	0
23	76	21	11		0	0	0
24	86	20	10		0	0	0
25	79	17	9		0	0	0
26	93	29	15		0	0	0

27	132	34	18	0	0	0
28	100	38	20	0	0	0
29	76	21	11	0	0	0
30	54	15	8	0	0	0
31	109	30	15	4	0	0
32	101	21	11	4	0	0
33	59	18	9	0	0	0
34	98	26	13	8	3	1
35	76	22	11	1	0	0
36	87	18	9	4	1	1
37	100	18	9	4	0	0
38	90	18	9	0	0	0
39	84	19	10	0	0	0
40	90	16	8	0	0	0
41	104	24	12	4	6	2.75
42	100	22	11	3	5	3.25
43	95	23	12	0	0	0
44	78	27	14	0	0	0
45	83	19	10	0	0	0
46	71	21	11	0	0	0
47	95	24	12	0	0	0
48	96	20	11	3	0	0
49	86	20	10	0	0	0
50	112	17	9	8	0	0
51	84	20	10	0	0	0
52	110	19	10	4	0	0
53	111	11	8	4	0	0
54	97	26	13	0	0	0
55	71	17	9	0	0	0
56	62	26	13	0	0	0
57	82	20	10	0	0	0
58	88	19	10	0	0	0
59	86	18	9	0	0	0
60	77	14	7	0	0	0
61	80	18	9	0	0	0
62	70	14	7	0	0	0
63	113	13	7	9	0	0
64	101	10	7	4	0	0
65	100	26	13	0	0	0
66	98	7	7	4	0	0
67	113	14	7	5	1	1
68	90	20	10	0	0	0
69	111	17	9	2	4	2
70	97	14	7	0	6	3
71	86	14	7	1	0	0
72	100	16	8	4	0	0
73	72	17	9	0	0	0
74	66	17	9	0	0	0

75	74	18	9		0	0	0

September 04-05 – Essex:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	88	25	13	0	1	0	0
2	87	20	11		3	1	0
3	74	24	12		0	0	0
4	67	17	9		0	0	0
5	61	20	11		0	0	0
6	98	20	12		0	11	1
7	87	25	13		0	10	1
8	84	27	14		0	15	1
9	77	26	13		0	14	1
10	90	24	12		1	14	1
11	69	17	10		0	17	2
12	92	23	12		0	16	1
13	79	23	12		0	16	2
14	83	26	13		0	0	0
15	93	20	11		1	0	0
16	100	8	8		4	0	0
17	67	15	9		0	0	0
18	80	25	13		0	0	0
19	74	28	14		0	0	0
20	79	17	9		0	17	2.4
21	131	20	11		3	0	0
22	149	27	14		6	6	1
23	129	18	10		3	2	1
24	100	20	10		3	7	1.3
25	112	21	11		4	21	1.36
26	79	23	12		0	22	1.63
27	98	22	12		3	17	2.25
28	84	25	14		0	22	1.5
29	59	17	9		0	0	0
30	70	16	8		0	15	1.625
31	64	21	11		0	0	0
32	74	12	6		0	12	2.1
33	65	20	10		0	0	0
34	66	20	11		0	0	0
35	54	20	11		0	0	0
36	71	6	4		3	0	0
37	73	14	7		2	0	0
38	85	38	19		0	0	0
39	74	24	12		0	0	0
40	91	6	4		4	0	0
41	72	20	10		0	2	1
42	84	15	8		4	11	1
43	79	17	10		3	8	1.2
44	65	16	8		0	0	0
45	98	0	0		8	0	0
46	83	20	11		2	0	0

47	98	20	10		3	20	1.5
48	90	21	11		3	1	1
49	102	21	11		5	21	1.3
50	63	13	8		0	0	0
51	123	20	13		10	16	2
52	96	22	12		2	21	1.6
53	58	12	7		0	12	3.3
54	120	18	10		5	17	1.3
55	76	20	10		0	5	1.3
56	59	20	11		0	20	1.7
57	90	20	10		2	1	1
58	74	18	9		0	2	1
59	82	19	10		4	6	1
60	83	19	11		2	2	1
61	107	19	11		2	12	1.5
62	58	16	9		0	15	1
63	71	18	10		1	6	1
64	60	19	10		0	9	1
65	76	15	8		4	12	1.5
66	108	20	11		2	0	1.8
67	99	23	12		5	23	1
68	81	26	13		0	18	0
69	71	16	9		1	0	0
70	78	21	11		0	0	0
71	97	15	8		3	0	0
72	110	16	9		6	7	1
73	100	17	10		4	9	1
74	99	17	10		2	1	1
75	80	18	10		0	0	0

September 04-05 – Chatham:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	79	13	7	0	1	0	0
2	66	11	6		0	0	0
3	107	14	8		3	0	0
4	86	16	9		1	0	0
5	94	15	8		0	0	0
6	73	14	8		0	0	0
7	87	10	5		0	0	0
8	65	12	7		0	0	0
9	63	15	8		0	0	0
10	56	8	5		0	0	0
11	63	7	4		0	0	0
12	89	10	8		0	0	0
13	61	24	14		0	22	1.9
14	90	18	10		0	11	1.8
15	64	17	10		0	4	1.75
16	66	6	5		0	0	0
17	79	1	0		0	0	0
18	61	4	5		0	0	0

19	75	4	3	0	1	1
20	51	5	4	0	5	2
21	50	6	5	0	6	2.5
22	72	8	6	0	0	0
23	110	18	10	1	0	0
24	120	12	8	3	0	0
25	129	5	3	2	0	0
26	115	6	5	3	0	0
27	83	16	8	0	0	0
28	80	25	14	0	24	2.1
29	50	17	9	0	17	1.2
30	55	17	9	0	17	1.2
31	86	14	9	0	14	3.4
32	100	21	12	0	0	0
33	71	19	11	0	1	1
34	82	12	10	0	1	2
35	67	11	6	0	0	0
36	91	24	13	0	0	0
37	95	28	14	0	0	0
38	99	26	13	0	0	0
39	90	24	12	0	0	0
40	98	24	12	0	0	0
41	65	21	11	0	0	0
42	71	24	12	0	0	0
43	114	33	17	0	0	0
44	71	16	9	0	0	0
45	102	30	16	0	0	0
46	80	22	11	0	0	0
47	110	15	11	0	0	0
48	67	14	8	0	0	0
49	85	9	5	0	0	0
50	100	19	10	0	0	0
51	84	12	7	0	0	0
52	83	14	7	0	0	0
53	70	17	9	0	0	0
54	73	17	9	0	0	0
55	73	23	12	0	12	1.7
56	104	23	12	0	23	4.4
57	75	14	8	0	14	4.1
58	77	20	11	0	20	3
59	100	25	13	0	25	4.75
60	97	21	11	0	21	3.3
61	97	20	10	0	8	3.4
62	61	20	10	0	0	0
63	113	23	12	5	0	0
64	117	21	11	0	0	0
65	124	17	9	0	0	0
66	159	15	10	3	1	2
67	88	19	10	0	0	0
68	79	15	8	0	0	0
69	114	23	12	0	0	0

70	69	17	9		0	0	0
71	66	18	10		0	0	0
72	100	18	11		0	0	0
73	115	22	12		0	0	0
74	100	17	9		0	10	3.2
75	63	20	10		0	13	3.5

September 04-05 – Port Stanley:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	82	9	6	0	0	0	0
2	75	10	6		0	0	0
3	110	14	7		0	14	0
4	98	18	10		3	16	1.14
5	100	8	5		0	0	2
6	99	17	9		0	0	0
7	110	13	7		0	0	0
8	104	15	8		0	0	0
9	98	12	7		0	0	0
10	100	12	6		0	0	0
11	107	11	6		2	0	0
12	95	4	3		0	0	0
13	100	9	5		0	0	0
14	110	14	8		1	1	1
15	114	16	8		1	1	1
16	100	11	6		0	0	0
17	109	14	8		3	2	1
18	107	12	6		3	1	0
19	110	11	6		2	2	1
20	93	12	6		0	0	0
21	93	12	6		1	0	0
22	85	16	8		1	0	0
23	79	8	5		0	0	0
24	98	11	6		0	0	0
25	89	9	6		1	1	1
26	85	15	8		0	2	1
27	91	14	9		0	0	0
28	100	11	6		1	0	0
29	97	12	7		0	0	0
30	84	10	6		4	0	0
31	60	5	5		0	0	0
32	61	8	5		0	0	0
33	60	5	3		0	0	0
34	120	18	11		0	0	0
35	95	0	0		6	0	0
36	98	0	0		3	0	0
37	95	0	0		3	0	0
38	108	0	0		3	0	0
39	127	0	0		7	0	0
40	96	0	0		1	0	0
41	100	0	0		2	0	0

42	130	0	0		3	0	0
43	100	12	7		0	12	1.75
44	106	14	8		0	7	1.25
45	77	16	8		0	0	0
46	83	16	8		0	0	0
47	60	11	7		0	2	1
48	75	11	8		0	0	0
49	68	10	5		0	2	1.5
50	83	22	11		0	0	0
51	98	7	6		0	7	2.6
52	101	0	0		4	0	0
53	123	10	8		1	5	1.75
54	123	18	11		0	14	2.3
55	110	9	7		0	8	1.8
56	120	9	6		0	8	2
57	144	15	9		4	9	1.71
58	118	12	7		0	2	1
59	97	14	9		0	3	1
60	119	14	8		0	5	1.4
61	102	4	3		0	4	3.6
62	126	16	8		0	0	0
63	115	12	9		0	0	0
64	112	18	9		0	0	0
65	126	0	0		4	0	0
66	119	17	10		2	17	1.5
67	137	0	0		0	0	0
68	125	15	9		0	0	0
69	140	16	9		0	0	0
70	134	21	11		0	0	0
71	145	20	11		0	0	0
72	141	18	10		0	0	0
73	133	20	11		0	0	0
74	112	18	9		0	0	0
75	95	17	9		0	0	0

September 04-05 – Simcoe:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	68	10	5	0	0	8	1.25
2	71	10	5		0	8	1.5
3	63	17	9		0	15	1.16
4	109	20	11		6	3	2
5	128	15	9		9	6	1.6
6	80	22	11		0	20	2.4
7	64	18	10		0	16	1.5
8	63	16	9		0	16	1.75
9	66	19	10		0	17	2.3
10	60	22	11		0	20	2.1
11	74	23	12		0	12	1
12	88	23	12		0	23	1.58
13	74	20	11		0	12	1

14	70	19	10	0	10	1
15	108	17	9	7	17	2.4
16	131	16	8	13	5	1
17	138	19	11	9	6	1.8
18	90	14	8	0	8	1.5
19	79	12	7	0	2	1
20	137	21	11	11	6	1.25
21	136	19	10	5	2	1
22	89	20	9	6	14	2.2
23	95	27	12	0	2	1
24	132	19	9	8	4	1
25	100	14	8	4	0	0
26	118	16	9	1	4	1
27	104	18	10	5	4	1
28	91	21	11	1	0	0
29	98	23	12	1	0	0
30	73	15	8	0	0	0
31	115	13	9	5	3	1.6
32	98	16	8	3	5	1.5
33	95	15	9	6	6	1.6
34	99	19	10	2	0	0
35	120	21	10	9	2	2
36	78	20	10	0	0	0
37	76	23	11	0	2	1
38	85	23	9	0	1	1
39	119	23	12	8	3	2.3
40	95	20	9	0	12	1
41	100	9	5	9	9	1.4
42	72	14	7	0	0	0
43	61	16	8	0	6	1
44	89	17	10	5	9	1.4
45	110	18	10	7	14	1.4
46	74	13	7	0	0	0
47	82	12	8	3	3	2
48	100	17	10	10	10	1.8
49	75	17	8	0	0	0
50	118	20	13	11	17	1.6
51	124	24	12	17	11	1
52	126	18	11	9	11	1
53	102	8	6	4	3	1
54	100	21	12	5	10	1
55	115	21	11	10	19	2.2
56	94	11	6	6	2	2
57	93	19	10	3	0	0
58	122	18	10	10	6	1.5
59	110	15	7	8	5	1
60	92	19	10	5	6	1.25
61	76	15	8	0	0	0
62	128	12	8	9	10	1.42
63	119	20	12	12	11	1.25
64	60	17	9	0	0	0

65	95	19	10		0	11	1.33
66	95	19	10		0	0	0
67	100	24	12		0	0	0
68	87	16	9		0	0	0
69	111	18	11		0	0	0
70	102	15	8		0	0	0
71	82	14	8		0	0	0
72	108	17	9		0	0	0
73	78	13	7		0	0	0
74	78	13	7		0	0	0
75	79	18	9		0	0	0

September 10-11 – Brantford:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	83	14	7	0	0	14	3.28
2	57	15	8		0	4	1
3	51	11	6		0	9	1
4	69	11	6		0	9	1
5	110	15	8		3	0	0
6	69	8	4		0	0	1
7	90	7	4		0	0	0
8	102	9	5		2	1	1
9	83	12	6		3	0	0
10	79	7	4		1	3	1
11	74	3	3		0	1	1
12	70	3	2		0	0	0
13	109	18	9		2	0	0
14	122	11	6		3	3	1
15	125	8	6		3	1	1
16	128	9	6		0	9	1
17	137	12	7		6	9	1
18	115	14	7		6	1	1
19	144	3	3		3	0	0
20	89	14	8		0	0	0
21	105	18	8		1	15	3
22	122	16	8		0	0	3
23	100	12	6		0	4	1
24	99	0	0		2	0	0
25	84	15	8		0	0	0
26	70	19	10		0	1	1
27	74	16	9		0	6	1
28	73	2	2		0	0	0
29	53	14	7		0	3	1
30	85	12	6		0	12	1.3
31	112	16	9		1	5	1
32	91	10	6		0	4	1.25
33	76	15	9		0	0	0
34	97	11	8		1	0	0
35	85	11	7		3	0	0
36	91	16	8		1	0	0

37	72	17	9		1	0	0
38	85	22	11		0	0	0
39	55	10	7		0	0	0
40	66	14	7		0	0	0
41	75	14	7		0	0	0
42	114	26	13		4	2	1
43	52	17	9		0	0	0
44	97	8	6		0	1	1
45	117	9	5		0	0	0
46	58	10	5		0	0	0
47	66	19	10		0	0	0
48	79	19	10		0	0	0
49	80	19	10		0	0	0
50	74	14	7		1	0	0
51	112	15	9		3	1	1
52	98	15	9		3	3	1.3
53	117	23	11		2	1	2
54	80	19	10		0	0	0
55	74	17	9		0	0	0
56	103	18	11		5	0	0
57	75	10	6		0	0	0
58	56	8	5		0	0	0
59	57	6	3		0	0	0
60	94	13	7		1	4	1
61	68	12	7		3	3	1
62	65	10	6		0	3	1
63	120	19	12		5	7	1.16
64	51	13	7		0	0	0
65	100	7	7		1	0	0
66	88	12	8		1	3	1
67	74	5	3		0	5	1.3
68	91	7	4		0	0	0
69	108	11	6		2	0	0
70	104	11	8		0	0	0
71	99	8	5		0	1	2
72	81	4	3		0	1	2
73	64	14	8		0	0	0
74	100	10	8		3	10	1
75	95	12	7		3	3	2

September 10-11 – London:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	103	22	12	0	3	4	1.3
2	63	15	7		0	6	1
3	74	16	8		0	1	1
4	83	14	7		0	0	0
5	101	12	8		5	4	1.5
6	80	17	9		0	0	0
7	85	5	4		1	1	1
8	81	13	7		1	0	0

9	72	16	8	0	0	0
10	86	18	9	0	0	0
11	79	6	4	0	0	0
12	100	14	9	2	0	0
13	95	11	6	5	4	1
14	76	21	11	0	6	1
15	124	15	10	4	3	1
16	121	20	9	4	1	1
17	100	13	7	1	1	1
18	130	8	5	10	3	1
19	81	17	9	0	0	0
20	78	11	6	0	0	0
21	85	9	6	0	0	0
22	100	18	9	1	2	1
23	96	13	7	1	0	0
24	108	19	9	3	3	1
25	115	25	11	0	0	0
26	94	19	9	2	2	1
27	99	2	2	1	0	0
28	84	20	9	1	2	1
29	83	8	5	0	0	0
30	68	17	9	0	0	0
31	88	19	10	0	1	1
32	72	11	7	0	1	1
33	84	18	9	0	0	0
34	80	20	10	0	2	1
35	79	18	9	1	3	1
36	86	20	11	0	0	0
37	79	4	4	0	0	0
38	88	12	7	0	0	0
39	65	11	6	0	4	1
40	78	13	7	0	1	1
41	97	14	8	0	0	0
42	95	8	6	1	0	0
43	107	25	13	1	6	1
44	114	7	5	8	1	1
45	110	22	11	2	8	1
46	89	20	9	0	0	0
47	91	13	9	0	1	1
48	100	26	14	0	6	1.75
49	115	25	13	0	1	1
50	112	26	13	0	0	0
51	101	20	11	0	0	0
52	97	27	14	0	0	0
53	117	22	11	0	1	1
54	133	23	14	2	1	1
55	105	22	12	5	3	1.3
56	130	19	11	2	3	1.67
57	137	27	14	0	8	1
58	103	7	5	0	0	0
59	123	11	8	8	10	1.4

60	74	10	6		0	0	0
61	77	20	10		0	0	0
62	82	15	8		0	0	0
63	104	19	10		0	0	0
64	74	20	11		0	0	0
65	80	11	7		0	0	0
66	84	16	9		0	5	1.3
67	83	8	5		0	0	0
68	152	29	15		8	4	3
69	94	27	14		0	0	0
70	99	22	12		0	0	0
71	73	10	7		0	0	0
72	94	14	8		0	0	0
73	79	22	11		0	0	0
74	100	22	11		0	0	0
75	100	14	7		0	0	0

September 10-11 – Grand Bend:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	99	26	13	0	0	2	1
2	79	22	11		0	0	0
3	88	12	7		0	0	0
4	79	24	12		0	23	1.09
5	66	18	9		0	0	0
6	95	23	12		0	22	1.9
7	88	17	12		0	10	1.16
8	78	18	9		0	15	2.63
9	83	16	8		2	15	2.5
10	50	8	6		0	6	2.4
11	95	13	7		2	9	2
12	122	21	11		3	21	1.7
13	124	18	9		2	16	1.89
14	127	24	12		0	22	3.54
15	100	26	13		0	13	1
16	82	18	9		0	0	0
17	120	20	10		6	8	1
18	83	20	10		0	2	1
19	86	21	11		0	8	4
20	61	10	5		0	8	1
21	52	10	5		0	10	1
22	61	13	7		0	5	2.67
23	81	24	13		0	4	1.3
24	67	22	11		0	11	3.4
25	92	16	9		3	16	1
26	99	16	9		1	12	1
27	106	22	14		0	18	1
28	77	15	8		0	2	1
29	81	14	7		0	8	1
30	84	19	11		0	19	1.18
31	102	14	9		1	14	1

32	84	12	8		2	9	1
33	88	16	8		0	10	1
34	79	9	5		5	9	1.6
35	87	16	8		8	11	2
36	85	18	9		9	0	0
37	108	15	8		8	6	1
38	129	12	6		6	11	1
39	70	10	6		6	10	1.3
40	113	28	14		14	16	2.16
41	88	15	9		9	15	2.875
42	68	9	6		6	9	3.2
43	95	20	12		12	15	2
44	98	22	13		13	11	1.33
45	91	20	11		2	1	1
46	100	24	12		1	0	0
47	71	20	10		0	0	0
48	97	20	10		0	0	0
49	110	14	8		0	0	0
50	130	22	11		5	0	0
51	117	19	10		2	0	0
52	91	20	10		0	0	0
53	91	19	10		0	0	0
54	115	18	10		0	0	0
55	125	18	9		3	0	0
56	117	16	8		0	0	0
57	92	13	7		0	0	0
58	100	12	7		0	0	0
59	100	17	9		0	0	0
60	100	14	8		0	0	0
61	113	15	8		1	0	0
62	123	19	10		0	0	0
63	113	14	8		0	0	0
64	119	13	7		1	0	0
65	130	15	6		2	0	0
66	123	19	10		0	0	0
67	122	23	12		0	0	0
68	129	16	8		2	0	0
69	126	23	12		0	0	0
70	115	20	10		0	0	0
71	100	17	8		0	0	0
72	116	21	11		0	0	0
73	130	20	10		3	0	0
74	140	9	5		7	0	0
75	136	18	9		3	0	0

September 10-11 – Sarnia:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	64	22	11	0	1	0	0
2	99	26	14		2	0	0
3	74	20	10		1	0	0

4	74	24	12	2	0	0
5	102	26	13	7	2	1
6	98	26	13	6	2	1
7	84	21	11	2	0	0
8	94	17	8	2	1	1
9	101	23	12	6	2	1.5
10	108	30	15	3	3	1.3
11	127	28	15	6	2	1.5
12	132	25	13	10	1	2
13	98	18	10	4	1	2
14	109	16	8	4	0	0
15	78	24	12	0	0	0
16	90	25	12	0	0	0
17	98	32	16	0	0	0
18	86	26	13	0	0	0
19	72	19	10	0	0	0
20	86	20	10	1	0	0
21	96	24	12	2	0	0
22	70	19	10	0	0	0
23	74	20	10	0	0	0
24	81	23	12	0	0	0
25	78	14	10	0	0	0
26	93	28	15	0	0	0
27	123	35	19	1	0	0
28	100	39	20	0	0	0
29	68	19	9	0	0	0
30	50	13	7	0	0	0
31	58	15	8	0	0	0
32	108	28	14	4	0	0
33	99	29	15	0	0	0
34	102	21	11	4	0	0
35	56	18	9	0	0	0
36	97	20	10	8	2	1
37	58	17	9	0	0	0
38	98	25	13	4	3	1
39	107	23	12	6	0	0
40	88	19	10	4	0	0
41	73	23	12	1	0	0
42	99	22	11	4	1	1
43	69	17	9	0	1	1
44	72	16	8	0	0	0
45	75	20	10	0	0	0
46	97	14	7	0	14	3.2
47	90	14	7	1	0	0
48	78	19	10	0	1	1
49	69	19	10	0	1	1
50	87	19	10	0	1	1
51	95	18	9	0	2	1.5
52	81	14	7	0	3	1
53	96	16	9	0	0	0
54	96	23	12	0	1	1

55	105	24	12		4	9	1.8
56	105	20	11		3	12	1.44
57	94	25	13		0	3	1
58	76	17	9		0	3	1
59	78	23	12		0	0	0
60	93	19	10		1	0	0
61	79	18	9		0	1	1
62	86	19	10		0	0	0
63	113	16	9		8	0	0
64	107	17	9		0	1	1
65	98	18	10		4	1	1
66	91	19	10		0	0	0
67	99	24	12		0	0	0
68	100	11	7		4	0	0
69	110	14	9		8	1	1
70	83	18	9		0	0	0
71	80	17	9		0	0	0
72	80	200	10		0	0	0
73	70	14	7		0	0	0
74	100	8	8		3	0	0
75	111	15	10		5	0	0

September 10-11 – Essex:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	64	21	11	0	1	9	1.4
2	74	20	10		2	5	1.8
3	77	22	12		0	12	1.12
4	60	22	11		0	1	1
5	61	22	11		0	6	1
6	73	19	10		0	3	1
7	100	22	12		3	1	1
8	71	9	5		0	9	2.4
9	130	24	12		3	24	1.3
10	100	15	10		6	0	2.6
11	94	17	11		3	17	2
12	100	4	3		6	4	1.46
13	85	14	10		2	14	2.8
14	93	21	12		2	21	1.42
15	74	20	10		0	18	2.22
16	58	16	8		0	2	1
17	59	19	10		0	7	1
18	62	15	8		0	7	1
19	77	22	11		0	8	1
20	77	24	13		0	19	1.4
21	89	21	12		2	11	1
22	70	22	12		0	8	1.75
23	96	21.	11		2	17	1
24	123	23	14		7	22	1.84
25	70	25	13		7	20	1
26	98	20	11		2	13	1.5

27	62	22	12	0	20	1.9
28	73	26	13	0	12	1
29	100	24	12	4	22	1
30	79	20	10	0	0	1
31	95	17	9	3	15	1.12
32	78	20	10	2	20	1.1
33	56	22	11	0	16	1
34	74	20	11	1	10	1
35	92	30	15	0	14	1.23
36	99	25	13	2	16	1
37	128	20	11	3	18	1
38	152	28	15	0	18	1.2
39	120	29	15	5	15	1.12
40	100	31	16	0	1	1
41	80	22	11	0	14	1
42	67	17	9	0	11	1.16
43	66	16	8	0	8	1
44	60	15	8	0	10	1.2
45	56	20	10	0	16	1
46	60	22	11	0	14	1
47	58	26	13	0	16	1
48	65	21	11	0	0	0
49	69	20	10	0	0	0
50	63	19	10	0	0	0
51	79	22	11	0	12	1.16
52	76	22	12	0	22	1
53	97	18	10	6	18	1.7
54	112	14	8	5	14	1.75
55	58	17	9	0	15	1
56	91	16	9	2	16	1.88
57	54	18	9	0	12	1
58	59	15	8	0	10	1
59	88	13	9	2	11	1.12
60	67	9	8	1	3	1
61	84	36	19	0	12	1.6
62	63	27	14	0	16	2.37
63	78	16	9	2	2	1
64	74	19	10	0	10	1
65	96	3	3	3	0	0
66	92	4	4	2	0	0
67	93	9	6	3	7	1
68	53	12	7	0	11	3.5
69	103	4	4	6	0	1.25
70	96	17	10	4	11	1
71	68	21	11	0	13	1
72	68	20	10	0	12	1
73	88	19	10	2	9	1
74	99	23	16	3	14	1.2
75	98	26	14	0	24	1.85

September 10-11 – Chatham:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	86	12	7	0	1	0	0
2	67	10	5		0	0	0
3	106	12	7		3	1	1
4	95	17	9		1	0	0
5	96	15	8		0	0	0
6	69	11	6		0	2	1
7	89	11	6		0	0	0
8	72	12	7		0	0	0
9	62	12	7		0	0	0
10	50	7	5		0	0	0
11	90	6	5		0	0	0
12	55	13	8		0	13	3
13	89	7	8		0	7	2.71
14	76	10	9		0	8	2.67
15	63	7	6		0	2	2
16	77	0	0		0	0	0
17	79	0	0		0	0	0
18	50	0	0		0	0	0
19	62	0	0		0	0	0
20	50	0	0		0	0	0
21	70	0	0		0	0	0
22	72	8	6		0	0	0
23	108	18	9		1	0	0
24	117	12	8		3	0	0
25	113	0	0		2	0	0
26	116	2	2		3	0	0
27	81	4	4		0	0	0
28	82	27	14		0	24	2.3
29	47	18	9		0	18	2
30	56	14	10		0	14	1.3
31	77	10	6		0	10	4.3
32	66	4	3		0	1	2
33	93	17	9		0	4	2
34	68	11	6		0	0	0
35	91	24	13		0	1	1
36	98	26	13		0	0	0
37	97	27	14		0	0	0
38	90	24	12		0	0	0
39	93	26	13		0	0	0
40	64	20	11		0	0	0
41	74	25	13		0	0	0
42	70	19	10		0	0	0
43	115	19	10		2	0	0
44	111	32	16		0	0	0
45	72	16	9		0	0	0
46	82	22	11		0	0	0
47	110	28	15		0	0	0
48	94	26	13		0	0	0
49	63	12	7		0	0	0

50	86	10	8		0	1	3
51	86	7	4		0	0	0
52	92	14	8		0	0	0
53	88	14	7		0	0	0
54	78	16	9		0	0	0
55	63	10	9		0	0	0
56	64	13	9		0	0	0
57	73	22	10		0	11	2
58	74	11	7		0	11	4.5
59	100	24	12		0	24	5
60	81	18	9		0	18	3.7
61	100	26	13		0	26	4.86
62	94	22	11		0	22	4
63	100	18	9		0	16	2.75
64	98	17	9		0	11	4
65	66	20	10		0	16	3.75
66	100	20	10		0	14	4.14
67	100	16	8		0	12	4
68	61	20	10		0	0	0
69	121	22	11		5	0	0
70	114	22	11		0	0	0
71	144	16	8		0	4	1
72	84	18	9		0	3	1
73	74	15	8		0	0	0
74	116	20	11		0	0	0
75	69	16	8		0	0	0

September 10-11 – Port Stanley:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	100	13	7	0	3	13	1
2	98	8	5		0	0	0
3	95	17	9		0	0	0
4	111	16	8		0	1	1
5	99	16	8		0	3	1
6	93	15	8		0	2	1
7	92	12	6		0	0	0
8	108	11	6		2	1	1
9	70	6	3		0	0	0
10	105	14	9		1	1	2
11	120	16	8		1	4	1
12	91	2	2		2	0	0
13	99	9	5		5	0	0
14	110	12	7		7	9	1
15	111	15	8		8	4	1
16	110	11	6		6	7	1
17	108	11	7		7	0	0
18	115	9	6		6	3	1
19	116	13	8		8	7	1
20	58	9	5		5	2	1
21	98	12	7		7	6	1
22	86	8	5		5	8	1

23	93	6	6	1	0	0
24	100	11	6	0	9	1.2
25	98	12	7	0	1	1
26	94	11	6	1	1	1
27	90	7	4	0	0	0
28	98	13	7	0	5	1
29	100	7	6	0	3	1
30	86	8	5	0	6	1.4
31	102	11	6	0	9	1
32	90	15	8	0	0	0
33	99	9	5	0	9	1
34	68	9	5	0	3	1
35	99	10	6	0	2	1
36	94	15	9	0	0	0
37	66	6	4	0	0	0
38	58	5	3	0	2	1
39	67	5	4	0	2	1
40	100	9	5	3	9	3.8
41	111	12	7	2	12	3.14
42	82	8	6	0	1	1
43	71	9	5	0	4	1
44	92	0	0	6	0	0
45	95	0	0	3	0	0
46	93	0	0	3	0	0
47	123	0	0	8	0	0
48	111	0	0	3	0	0
49	101	0	0	1	0	0
50	109	0	0	2	0	0
51	130	0	0	3	0	0
52	132	0	0	3	0	0
53	113	0	0	3	0	0
54	99	9	6	0	9	3.5
55	116	11	7	0	11	2.42
56	87	4	2	0	4	3
57	121	115	9	3	12	1.5
58	96	0	0	1	0	0
59	89	6	5	0	6	4.2
60	115	15	8	0	15	3.125
61	140	16	8	1	15	1.375
62	120	10	7	1	4	1
63	96	8	5	0	8	2.8
64	87	3	3	0	3	2.3
65	139	15	9	0	10	2
66	133	7	5	1	6	1.5
67	113	14	7	0	12	3
68	100	12	7	0	12	3.63
69	98	14	8	0	14	3.75
70	94	11	8	0	11	3.63
71	97	12	6	0	12	3.67
72	101	13	8	0	0	3.85
73	103	15	8	0	6	2.67

74	100	10	6		2	9	1.2
75	91	8	5		0	8	1.6

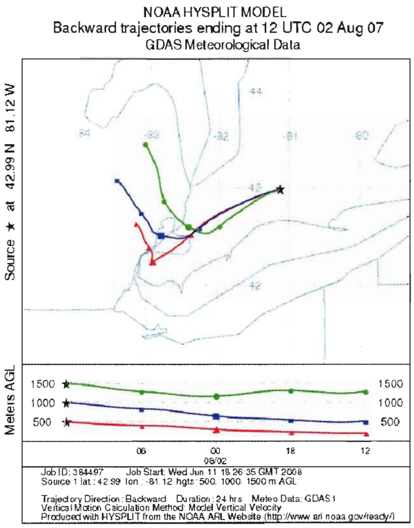
September 10-11 – Simcoe:

Plant #	height cm	no. leaves	no. pairs	no. flowers	no. pods	no. injured	avg. injured area (H-B)
1	92	20	10	0	0	20	1.8
2	97	23	12		0	11	1
3	63	16	9		0	14	2
4	58	22	12		0	0	3.3
5	50	20	10		0	16	1
6	65	12	6		0	8	2
7	64	14	8		0	10	1.6
8	88	22	11		0	22	2
9	82	22	11		0	14	1
10	78	16	8		0	16	1.75
11	98	20	11		0	19	1.2
12	71	17	9		0	15	1
13	111	20	11		6	4	1.3
14	172	16	9		0	7	1.42
15	106	18	10		6	3	1.3
16	118	19	10		0	8	1.5
17	134	20	10		12	7	2
18	109	17	9		0	17	2.25
19	131	15	8		12	15	1
20	137	19	10		9	7	2.2
21	144	20	10		11	6	1
22	139	18	9		5	1	2.2
23	122	20	11		0	14	1.37
24	114	16	10		4	6	1
25	100	14	8		4	0	0
26	69	10	5		0	0	0
27	110	18	10		6	6	1.25
28	97	18	11		0	0	0
29	97	23	12		0	0	0
30	77	15	8		0	0	0
31	104	13	7		5	2	2
32	100	16	8		3	3	2
33	100	15	10		6	3	1.67
34	100	19	11		2	2	1
35	100	10	5		9	10	2
36	71	16	8		0	3	1
37	94	18	10		5	10	1.14
38	78	11	8		0	4	1
39	92	15	8		0	0	0
40	81	13	7		0	1	1
41	97	17	11		9	16	2.2
42	89	12	8		3	1	0
43	99	16	10		10	14	1.375
44	74	15	7		0	4	1
45	121	17	10		9	2	1
46	116	15	9		9	15	1.11

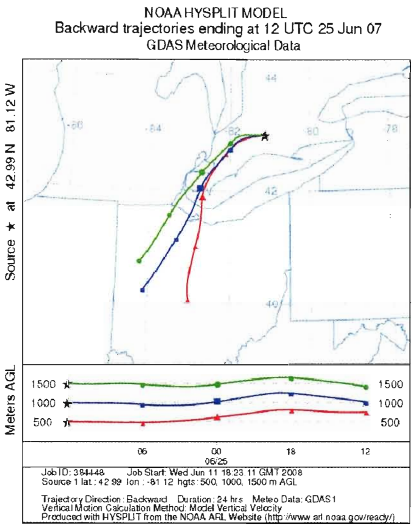
47	114	19	10	5	6	1.6
48	101	17	8	0	2	1
49	56	14	7	0	8	1
50	78	16	8	6	15	1
51	104	21	11	5	9	1.83
52	112	21	11	11	21	1.9
53	95	19	10	3	0	0
54	94	11	6	7	1	3
55	77	14	7	0	0	0
56	57	10	5	0	0	0
57	72	12	6	0	0	0
58	130	11	7	9	10	1.3
59	120	13	7	1	0	0
60	120	14	7	1	2	2
61	91	19	10	4	7	1.25
62	112	14	8	7	5	1
63	120	17	10	11	5	1
64	121	23	14	13	22	1.3
65	64	15	8	0	0	0
66	96	17	9	0	9	1.8
67	83	83	10	0	0	0
68	86	86	8	0	0	0
69	100	100	12	0	8	1
70	92	92	8	0	1	1
71	110	110	11	0	1	1
72	108	108	8	0	0	0
73	87	87	10	0	17	1.1
74	70	70	8	0	0	0
75	84	84	9	3	0	0

Appendix VII: Twenty-Four Hour Backwards Trajectories

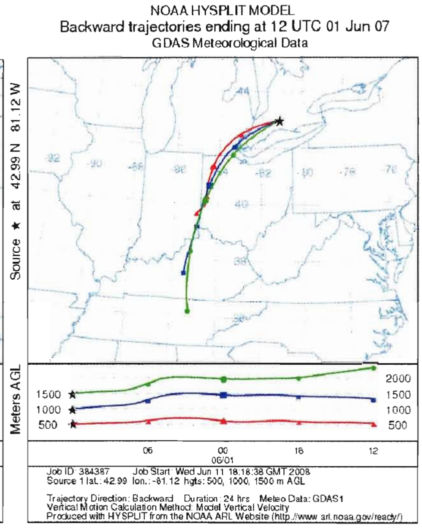
In order to determine where air parcels originated from on the three highest O_3 days in 2007, August 20, June 25, and June 07, backward trajectories were plotted that ended in the study region at 500, 1000 and 1500 meters above ground level. This was done using the National Oceanic and Atmospheric Administration (NOAA) HYSPLIT Model software with historical information retrieved from the global data assimilation system (GDAS), which was accessed on June 11, 2008 at www.arl.noaa.gov/ready/. In each image the star represents approximately London, Ontario and the end of the 24 hour trajectory.



A.



B.



C.